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THE

MICROSCOPE

IN

MEDICINE.

THE MICROSCOPE IN MEDICINE.

BY

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WITH MORE THAN 500 ILLUSTRATIONS, MOST OF WHICH HAVE BEEN DRAWN ON WOOD BY THE AUTHOR.

FOURTH EDITION,

MUCH ENLARGED.

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TO

MY FRIEND

HENRY W. ACLAND, M.D., LL.D., F.R.S.,

PRESIDENT OF THE MEDICAL COUNCIL,

REGIUS PROFESSOR OF MEDICINE IN THE UNIVERSITY OF OXFORD,

AND HON. PHYSICIAN TO H.R.H. THE PRINCE OF WALES.

PREFACE.

THE present volume, though larger than any of its predecessors, does not contain many things which might with propriety and advantage have been included. In it the author has recorded his experience of those methods of examining the tissues and fluids of the body in health and disease, which he has found most successful. He trusts the book will prove useful to those engaged in medical practice, and afford help and encouragement to students, who by devoting themselves to this branch of research hope some day to be able to add something to our knowledge of those marvellous phenomena upon which every action in health and disease depends. It is upon progress in this direction that we base our hopes of the more efficient prevention of disease, and the further alleviation of suffering.

61, GROSVENOR STREET,
October, 1877.

PREFACE

TO

THE FIRST EDITION.

A SHORT course of lectures, given in the spring of last year, forms the basis of the present volume. To the notes which had been prepared, and which the author had originally intended to print for the use of his pupils, much has since been added; and it is hoped that, in its present shape, the work may afford some assistance to practitioners and students in medicine who employ the microscope in clinical investigation, or in physiological and pathological inquiries.

In the present day, this branch of investigation is being pursued by all who are most anxious to increase our knowledge of the structural alterations taking place in disease, and of adding to our information concerning some of those important processes which interfere with the due performance of the healthy functions of different organs—investigations in which all may find ample employment, and may thus contribute to the advancement of the true interests of their profession, and aid in the elucidation of truths which may ultimately promote the interests and welfare of mankind in a degree not less than they will add to the advancement of science.

Except in cases referred to in the text, the woodcuts, which have been carefully executed by Mr. Davies, have been copied from drawings taken by the author from objects actually under observation.

In preparing the work, the author has to acknowledge the assistance he has derived from the suggestions of many; and he is very desirous of taking advantage of this opportunity of acknowledging how much he owes to his kind friends, Dr. Todd, Mr. Bowman, Dr. Johnson, and Dr. Acland, not only for the valuable assistance and information which he has on all occasions derived from their instruction and advice, but also for the warm encouragement they have constantly afforded him while he was a pupil, and ever since.

To his friend, Dr. Conway Evans, the thanks of the author are also due for much kind assistance.

27, CAREY STREET,
4th April, 1854.

PREFACE

TO

THE SECOND EDITION.

THE author has endeavoured to increase the usefulness of the work, and render it as practical as possible. With this view it has been revised throughout, and many of the articles have been entirely re-written. Much that related merely to manipulation in the first edition will be found in "How to Work with the Microscope," and has, therefore, been omitted in the present one, in place of this, much matter bearing more exclusively upon Medicine has been introduced, and upwards of sixty new and additional woodcuts have been inserted.

27, CAREY STREET,
October 1, 1858.

PREFACE

TO

THE THIRD EDITION.

THE continually increasing importance of minute microscopical inquiry to those engaged in investigating the nature of disease and promoting the advance of medicine, has rendered it expedient to improve this book, and make many additions to it. Although increased professional and other duties have prevented the author from devoting as much time to its revision as he could have wished, and the work is still far from what he desires it should be, the present edition is in many respects an improvement upon the last. The work now contains fifty-eight plates, which have been arranged and printed with the greatest care. The text has been revised throughout, and nearly 100 pages of new matter added. In order that these changes might be made without considerably increasing the size and price of the volume, and subjects of the greatest importance to the practitioner fully treated of, it has been necessary to omit the chapters on the structure of the healthy tissues and organs. This is of little importance, since this part of the subject is treated of in many other works, and much of what was published in previous editions will be introduced into an enlarged edition of the author's work on the tissues, upon which he is now engaged.

With the view of enabling the student to acquire, with as little trouble as possible, correct notions of the appearance of various objects, the number of illustrations in this work has been largely increased. Many of them have been drawn on wood or stone by the author himself. Some experience in medical teaching has convinced him that, in many cases, careful drawings may be substituted for long descriptions of objects with advantage. He feels that in these days, when there is so very much that must be learnt, it is the duty of the teacher to study not only how correct ideas may be conveyed to the student's mind, but how these may be communicated most simply, and most pleasantly.

The author's thanks are especially due to Mr. Sorby for kind assistance in revising the paragraphs upon spectrum microscopic analysis, to Mr. Lockhart Clarke for the directions for preparing specimens of the brain and spinal cord given on page 24, and to Dr. Tilbury Fox and Dr. Tonge for help in the preparation of the chapter on parasites.

Of the new drawings, some have been engraved by Miss Powell and others by Mr. Hart.

61, GROSVENOR STREET,
November, 1866.

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THIS work may be "read" by carefully studying the figures, and then referring to the text. A description of every drawing is placed beneath it, and, in most instances, a reference is given to the very page upon which the subject of the drawing is considered. A teaching experience, extending over more than five-and-twenty years, has convinced the author that, although the student will certainly obtain more correct views upon the microscopic characters of objects by attentively examining accurate representations, than by reading over and over again the most minute and elaborate descriptions of them, the information thus gained will be of little real use unless the student himself prepares and examines actual specimens, and makes careful drawings of what he sees.

THE

MICROSCOPE

IN

MEDICINE.

INTRODUCTORY.

A Plea for the Scientific Investigation of Disease.—The value of Scientific Researches, conducted by properly trained Members of the Medical Profession.—How new knowledge concerning the nature of Disease is to be acquired, and practical Medicine advanced.—Pathological Laboratories and Microscope Rooms in Public Hospitals.

ALTHOUGH the investigation of the nature of disease has been held by some to be distinct from its practical treatment and the relief of present suffering, it will now be generally admitted that scarcely any addition to our knowledge of disease can be made without leading sooner or later to an advance in practical medicine. Still by many benevolent persons no connection is believed to exist between the investigation of disease and the treatment of the sick. As a consequence of this doctrine, scarcely any efforts are made by wealthy men to advance the scientific investigation of disease, or to encourage efforts being made to discover means for its prevention, while millions have been spent upon the relief of present suffering. It is not to be wondered at that in a practical country like England there should be a general notion, that of the money contributed for the support of the sick poor during illness not a fraction should be devoted to scientific medical investigation, because no immediate benefit to the poor could be promised, and nothing tangible or intelligible to the public *might* be gained in place of the money that had been expended. Nor can anyone connected with hospitals be astonished that there should be a prejudice on the part of some of the generous supporters of hospitals

against minute and elaborate investigations. These people want the practical doctor, not the person who always has his eye at the end of a microscope, or is continually fishing for new facts in the excretions of the sick; and it is wonderful how many people who give money to medical charities have been persuaded, or have persuaded themselves, that no practical doctor can be a well-informed, thoughtful observer, and scientific investigator. Nevertheless, the opinion that the nature of disease should be thoroughly investigated seems to be gaining ground, and as sanitary information becomes diffused, it is to be hoped this idea will be so generally acquiesced in, as to be practically acted upon.

The want of real knowledge concerning the origin, nature, and properties of contagious poisons which have been so fatal in all ages, to man and to domestic animals, must have struck every intelligent person. Not only have earnest physicians of the days that have gone felt and acknowledged how little they knew, but the difficulties of the investigation in their time appeared insurmountable, while the means required for attempting such an inquiry were not obtainable. Many have in consequence been deterred from attempting it, although they were well qualified for the task.

But in these days the methods at our disposal for investigating such questions are far more numerous and efficient than was formerly the case, and they are becoming more and more perfect every day, while from time to time completely new modes of inquiry are discovered. The public may therefore reasonably expect that the important questions, concerning the nature, mode of transmission, and prevention of many diseases should be subjected to yet more searching and minute scientific investigation than has hitherto been practicable. It is, however, doubtful if many who hold this opinion have any accurate conception of what is involved in a thorough scientific inquiry. The practical object of undertaking such an inquiry is undoubtedly to discover a method for preventing or curing the malady, and this great end can be fully appreciated by every one; but yet it is certain that laborious and life-long investigations must be prosecuted before a result so much desired is rendered possible. The work would have to be carried on unremittingly, even though there might be little hope of immediate success.

Not a few scientific researches will appear to many persons to have been undertaken without any object whatever, and the time spent upon them no better than wasted. Nay, the greatest *scientific* successes in many departments of work have been regarded as worthless by men who considered they possessed a very large share of common sense, simply because they did not lead to immediate practical results, or demonstrable benefits at the time. And it must be freely admitted that every disease that we are acquainted with, and especially all contagious diseases, including those of animals, have been studied over and over

again by most excellent observers and thoroughly practical persons, for the very important object of discovering a remedy and for diminishing the ravages of such maladies, and hitherto, it must, I regret to say, be confessed that with a few remarkable exceptions, only very moderate success has been attained. Still there is enough to encourage us to proceed, and we may feel sure that however little may yet have been gained, new inquiries will certainly teach us more, and that by degrees the knowledge every generation of medical practitioners has longed to possess, will be obtained.

Scientific inquiries of the kind referred to must be undertaken by persons who are ready to work on in a thoroughly scientific spirit, for it is certain that the prospect of discovering anything practically useful is so small in proportion to the thought and labour and money that must be expended in the investigation, that scientific investigation is not likely to be taken up—and if taken up would not be patiently prosecuted to any useful conclusion—by those who work merely for notoriety, or by those who work merely for pay. Such an investigation must be conducted in a spirit of *pure inquiry*, and he who undertakes it should do so with the determination to study and to work, conscious that all his study and all his work may be without practical, or, in the ordinary sense, useful results. But, although some will smile at the idea, there are men who really love study, and who love work for the sake of work and study, and for the mental pleasure to be derived from work and study, and not alone for the rewards that even the greatest success may bring In many instances the hope that the inquiry itself, although leading to no useful results, may stimulate others to work on beyond the point arrived at, or the conviction that practical results will follow after many years, is a stimulus sufficient to excite some men to prosecute laborious research. But it is scarcely worth while discussing what particular motives men may have in pursuing unremunerative work. It is sufficient that the work should be done. Real advance is made by those who work for work's own sake, who desire to live that they may be able to work, and not by those who are obliged to work that they may live, or by those who work only in order that they may live easily; but unfortunately the number of men qualified who are able to work without pay is very small.

Much labour and money have been spent in obtaining statistical information concerning many diseases. Careful obvervations of a general kind have been prosecuted, and doubtless thoroughly well prosecuted, and accurately recorded for Government, and an enormous amount of information has been published in Blue books. But why can we not have, in addition to this, the results of very minute and careful investigations by physicians who have made themselves skilled physicists, chemists, and microscopists?

But it may truly be said that scientific men have already been called upon to undertake scientific work for several departments of Government and for Royal Commissions, but the sums awarded on few occasions for purely scientific work have been so very small, that it seems probable that those who advised the expenditure were either quite ignorant of the nature of the work to be undertaken and of the time it would occupy, or were careless whether Government and the public were properly informed of the advantages likely to result from the continued and thorough prosecution of such work, or felt that to ask for proper support would be injudicious and would be met by a blank point refusal. I cannot, however, but think that if attention had been properly directed to the real importance of the scientific investigation of disease, the public would have long since desired that investigations should have been systematically entered upon, and continuously prosecuted. Instead of the matter being openly explained, the propriety of obtaining the paid services of scientific men has been so timidly if not apologetically suggested, that it would almost seem as if those who advocated and recommended scientific work were doubtful if they were doing right, or were afraid of being laughed at by their political friends or opponents.

It may be an absurd suggestion, but to me it seems a most reasonable one, that in such a country as ours there should be scientific advisers appointed from time to time by the Government,—men who should hold office for *short periods only*, for in many branches of science, progress is so rapid that the aspect is entirely changed every few years, and in some departments a man who was an adept twenty years ago, might not even know how to use instruments only recently invented. Moreover, some scientific men in authority do occasionally exhibit partiality for particular schools of thought and work, and it is very desirable that investigation should not be carried on by one school alone. Nor is it reasonable to expect that a man of fifty or sixty should take up an entirely new method of inquiry, especially where considerable practice was required to carry it out successfully. The energy and enthusiasm necessary to enable a man even to attempt a difficult, and to many persons a hopeless scientific task, are much more likely to be found in men of thirty than in men of fifty. These inquiries are just suited for the younger scientific practitioners in medicine to attempt, and this may be said without implying the slightest disparagement of the seniors.

Is it not time that the Government and the country should be alive to the real usefulness of scientific work for other purposes as well as for forging the most destructive instruments of war? And if in this country we succeed in producing the most effective engines for the destruction of human life, is it not possible that by ingenuity employed

in another direction we might ascertain exactly how life may be most efficiently preserved, and rendered most happy while it lasts? The effective prosecution of the latter very wide inquiry may be conducted, and with all the requisite minute details, for a fraction of the thousandth part of what is now spent, and rightly spent, upon the former.

The wide general bearing and possible practical advantage of many of the most minute and abstract scientific inquiries is as yet but little appreciated, and I may perhaps be permitted to give one or two illustrations which may help to make this evident.

We know, as yet, very little of the nature of muscular contraction, but this *scientific question* bears in a very important manner upon the practical question how to get the greatest amount of work out of the muscles, and at the same time to preserve them in a healthy state with the smallest expenditure of food. And many will be astonished at the sort of detailed inquiry which must be undertaken before we can hope to arrive at a thorough knowledge of this subject. It may be possible to ascertain the structure and mode of action of muscles in the lowest animals, while the methods of examination yet known do not enable us to do so in man and the higher animals. We may find by observation that from the fact of the elementary particles of the muscles being larger, or more distinct, in some one of the lower simpler creatures than in the higher animals, great facilities are thereby offered for the inquiry. A man may have to spend years upon the study of minute points in the organisation of some worm, in order to determine some general question of the highest importance to mankind, and this may be the only possible method of arriving at sound conclusions.

To take another instance. From various circumstances it may be impossible to follow the ultimate arrangement of the nerve fibres in any of the tissues of the higher animals, or to prove, conclusively, by any mode of experiment, the general plan of their distribution, while in some small organ of an insect, or in a transparent papilla upon the surface of some microscopic worm, or mollusk, it may be that a nerve may be followed directly from its centre to its peripheral distribution and back again to the centre. The positive knowledge thus gained, which may have taken months or years to acquire, and apparently so unimportant and useless, might lead almost immediately to the determination for once, and for ever, of the general arrangement of nerves. It might enable us to arrive very shortly at the mode of action of nerves, and the nature of nerve-force, and to explain the phenomena of very many nervous diseases now but imperfectly understood, and, perhaps, to far more efficient modes of treatment than any we are now acquainted with.

Such inquiries are often prosecuted for years without results, or only with imperfect results; but the examples I have adduced may serve

to show that certain minds may with reason and pleasure strive for what it may be, at least in their day, impossible to obtain; but they strive on, and in spite of failure, for they know that those who have achieved the greatest scientific successes have had to encounter and patiently bear the greatest number of failures. But it is surely well for mankind that there are men to work, and the work done by them ought to be respected, even though its bearing may not be clear, or its usefulness evident at the time it is performed.

Next, then, let me consider how and by whom the most thorough investigation for example of the nature of contagious diseases of animals and man can be made. It will be conceded that it is desirable in the first place to ascertain, if possible, the *nature* of the contagious poison; next to discover how this passes from the infected to the sound organism; next to ascertain by what channels it enters the body, and if it is shown that it exists in the blood, it is most important to ascertain the exact changes induced in the composition of that fluid, and the secondary alterations resulting therefrom in the various tissues and organs of the body.

Now it is obvious that for conducting such an inquiry as this several different methods of investigation are necessary. Extensive chemical examination must be undertaken with the view of ascertaining in what particulars the fluids and solid organs of the infected organism differ from those in a sound organism, and this at every period of the disease from the moment after the poison gained entrance to the body to the time when the life of the organism attacked is destroyed.

Careful and detailed microscopical examination should be prosecuted, for the purpose of discovering if any unusual particles existed in the diseased fluids, and for demonstrating the exact changes in structure which had taken place in the tissues. And there are very numerous other methods of physical research, which are also necessary, but to these I shall not allude here. The symptoms occurring during life should of course be accurately noted by persons well accustomed to medical observation, and the morbid appearances observed after death.

Importance of investigating the Diseases of Animals.—We have been, it seems to me, far too inattentive and careless concerning the investigation of the diseases of animals. In them we have a wonderful opportunity of studying the course of many disorders almost identical with some of those from which man himself suffers. While a vast number of diseases of the lower animals, though differing in important points from the ailments which affect humanity, exhibit, nevertheless, characters which compel us to regard them as being of the same class, and justify us in studying them and reasoning from the phenomena according to the same principles. No one could have watched the

course of that wonderfully fatal fever, cattle plague, without coming to the conclusion that he was studying a disease of the same class as that in which are comprised the special fevers of man, and one which destroyed life in the same sort of way as many of our fevers. We had the opportunity of studying this disease stage by stage, and I am sure everyone who engaged in the work under the Royal Commission regrets that he was unable to do more, and that better means were not taken, to gain still more minute results, and larger funds placed at his disposal when this disease was raging. The opportunity of further investigations upon this malady will, let us hope, never occur in this country again. Lord Spencer, the Lord Lieutenant of Ireland, made the following very interesting observations, in connection with this matter, at the meeting of the Royal Dublin Society's Cattle Show, April 15th, 1869 :—

"Now, I often regret that there is not more sympathy and union between what are commonly called the medical and veterinary sciences. I believe if there were more union between the medical and veterinary bodies there would be great mutual advantage. I believe that not only would the veterinary science gain immense advantage from the knowledge and experience of the higher profession, but I believe that great benefit would be derived to medical science by being able to make experiments on animals. I remember well when I had the honour of being on the Cattle Plague Commission, that we carried out several experiments with regard to the diseases of cattle. We had several distinguished medical gentlemen on the Commission, and, with their consent, we entrusted to the officers of the Commission the execution of several experiments on animals. Many curious experiments were made with regard to disinfection, with regard to inoculation with disease, and with regard to particular remedies ; and I believe very important discoveries were not only made for veterinary science, but also, I believe, for the treatment of the human race. Now, I will not point out—it would not be my place to do so—how this could be obtained, but I believe it would be of great advantage to this country that a good veterinary college, or some department in that line, should be formed in Ireland."

I am quite sure that many of us would have been only too glad of opportunities of carrying out such investigations. The difficulty is the means.

Here, then, in connection with investigations upon the diseases of domestic animals, is work for the prosecution of which the assistance of several different persons, skilled in many different departments of science, will be required. The desirableness of all this will doubtless be generally admitted, and it will probably occur to every one that such comprehensive inquiries should be set on foot by Government.

Now, even if the Government thought it right to devote large sums of money to the purpose, it is doubtful if this, the most expensive

would be the most advantageous method of conducting the inquiry, for everything would depend upon the person selected to be at the head. It is scarcely possible to find a man himself practically well versed in all the branches of investigation that would have to be undertaken, while it is obvious that there would be great practical difficulties in obtaining a staff of thorough scientific workers who would submit to carry out the ideas and follow the mandates of one who perhaps knew far less of the subject than the workers themselves. Nor is it reasonable to suppose that a skilled scientific observer would submit to have the precise course he was to pursue dictated by another. Success in laborious scientific investigation seems to be due mainly to the intensity of the energy and enthusiasm of the individual who undertakes it, and for a man to work at anything with energy and enthusiasm, it is necessary that he be perfectly free to prosecute his work in his own way, and at his own time. Almost all the great work of any kind that has ever been done has been done by those who have freely followed the course which they themselves struck out, and it is most unlikely that a single head or the most admirably chosen committee, whose conclusions might be but a series of compromises between conflicting views, would suggest the exact manner in which each scientific man could think and work to the greatest advantage.

Moreover, there is something inexpressibly disappointing in having the results of months of hard work and thought commented upon, in a few short sentences, by one or more who can have had no experience whatever in the matter, who have never done such work, who perhaps dislike it and even sneer at it. Injustice of this kind beyond measure has been done by those in authority towards those who have worked. To a mind truly scientific there is nothing so dear as independence. Success is purchased too dearly if loss of independence is (as it certainly is in many cases) the price to be paid for it.

If, therefore, scientific work is to progress in England, I believe it will have to be conducted without help from the State, unless that help can be given to persons without reference to their general scientific opinions. I can conceive nothing more difficult for a Chancellor of the Exchequer, ignorant of science, than to ascertain the real merits of young scientific men, or to find out how to select those who would most probably make the best and most original workers and thinkers. Many a thorough worker is hopelessly disappointed in early life. He may be intelligent, patient, energetic and careful, and may possess the most wonderful power of work, and yet he may be of too independent a disposition to please the master under whom he happens to serve, or he may be haughty and disagreeable and offend him, or he may not pay that deference which many scientific authorities, however disinclined to defer to others, not unfrequently exact from those who assist them.

The idea, therefore, of a large department with a skilled head, and skilled scientific men following out different branches of an extensive investigation, with one common purpose, is an idea, however perfect it may seem in theory, not likely to work well if carried into practice. The same objections apply and with almost equal force to any attempt being made by a number of private individuals, an association, a company, or a society.

No attempt is likely to succeed which does not leave each man free to work in his own way, according to his own views. Chemists, physicists, and microscopists would look at the same inquiry from very different points of view, and each would ascertain facts of a very different nature. The conclusions arrived at might all point in the same direction, or in many different directions. Much capable of immediate application might be gained from their work, or nothing which at the time threw any light upon the question they had been called upon to consider. But I think that, although there are some objections even to that mode of conducting a scientific inquiry, it is only by permitting each person engaged to work in his own free way, that any good results would be rendered possible. Of course he would be able to set assistants to work for him, but he alone would direct them. Any man who has carried out work according to the directions of another, and work as suggested by himself, knows how hard and wearisome is the first, and how light and pleasant the last. Every worker ought to describe the results of his work, and should write his own report. Such a man would probably express the conclusions he had arrived at, at least as clearly as anyone else could express them for him. There is probably no task more difficult than that of carrying out a prolonged and difficult scientific inquiry according to a scheme designed by another, and especially by one who does not himself take part in the actual work.

Should Scientific Inquiries be conducted by Practitioners?—As regards the question whether, for instance, the nature of contagious poisons and such like purely scientific inquiries should be practically carried out by men who devote themselves to scientific investigation only, or by members of the medical profession, or, in the case of a cattle disease, by these and veterinary surgeons conjointly:—I would remark that the subject is one in which all who are engaged in the prosecution of medical research are specially interested. Their practical acquaintance with disease would necessarily tend to make them try to study the question in the simplest and also in the most efficient manner possible. The lessons inevitably taught by the constant observation of disease, and the habits of thought acquired by constant association with sick persons, ought not to make men dogmatists, but rather earnest and thoughtful students, while nothing can be more likely to kindle in the mind an intense desire to arrive at the truth than the consciousness of

the little that is really known even of the nature of diseases which they are daily called upon to treat.

But it may be said, the business of professional life itself unfits the mind for purely scientific investigation, while the imperative engagements of one whose first duty is attendance on the sick so seriously interfere with the laborious prosecution of detailed scientific observation as to impair the value of any results obtained, if not to render them worthless; and it will be remarked that although every medical practitioner has had a scientific education, few in the profession have had an opportunity of pursuing scientific studies for a sufficient period of time to enable them to gain a thorough knowledge of, far less a practical acquaintance with, various details necessary for the useful prosecution of original research.

Again, it has been urged that the followers of medicine are led to put trust in methods of treatment, of the usefulness of which there is the greatest doubt; and that medical men resort to a number of fanciful hypotheses to explain phenomena which they cannot account for in any more satisfactory way, and act as if these hypotheses were actual truths; and hence that such persons are not very likely to arrive at the truth in a difficult and prolonged scientific inquiry. Of late years medical methods of inquiry have been seriously attacked, and many of our doctrines, formerly regarded sound, and acted upon as if true, have been actually proved by ourselves to have been founded upon false premises. The very first principles of our science have been greatly modified, and not a few distinguished physicists and chemists have asserted that our vague notions concerning the peculiarity, if not mystery, of vital actions, are false. But although we may plead guilty to some of the charges which have been brought against us, it cannot be said with justice that we have discarded or overlooked the revelations of modern physics and chemisty. On the contrary, many medical writers have accepted mechanical doctrines which have not been proved and have grounded upon them theories of disease which are opposed to facts well known to every one who has observed in the wards of a hospital. It has been said over and over again that all the wonderful phenomena familiar to us, both in healthy and diseased living beings, are due to the ordinary forces of matter, and to these alone. And of late the public has been led to suppose that the progress of physical science has been great indeed, but that medicine has been at a comparative standstill. The fact is generalisations concerning physical causation have been made upon most insufficient data and people have been led to look upon many statements as facts which are only very confident assertions, the truth of which has not been proved, and is, probably, not provable.

We would gladly welcome pure physicists and chemists to our hospitals and medical institutions, and thankful indeed should we be if

they were enabled to add to our knowledge of disease, but they must not try to force upon us a number of conjectural dogmas which really add nothing whatever to our knowledge.

It seems to me that there is an unanswerable argument in favour of scientific medical inquiries being undertaken by medical practitioners themselves, viz., that no other scientific men are likely to engage in them; for although they may condemn medical science, investigators in other departments of science will not leave their own special inquiries for the purpose of investigating questions purely medical, and quite unremunerative. The scientific work of the chemist and physicist receives direct and remunerative return in connection with the advance and development of arts and manufactures. But the investigation of disease is not a subject which the present generation cares for, because the return for the time and labour expended is distant and doubtful,—hardly to be looked for until those who have done the work shall have passed away.

If, then, we cannot expect to obtain the services of distinguished anatomists, physicists, and chemists to assist us in the investigation of disease, it is obvious that if the work is to be done, medical men must make themselves thoroughly acquainted with these branches of science. And indeed medical practitioners have already added far more to these branches of knowledge than those who devote themselves entirely to their prosecution have added to medicine. Formerly the chief scientific authorities were members of the medical profession. In the present day we may boast of many thorough scientific workers, but we want many more to engage in the investigation of medical scientific questions of the utmost importance to the public, which can be studied only by men who at the same time are thorough doctors, and thorough men of science. And in what way, I would ask, can science be turned to better account practically than in investigating the nature of disease?—for every one who has thought at all upon the matter is probably thoroughly anxious that our knowledge of medicine should be extended, because common sense tells him that the more thoroughly we are acquainted with the exact nature of disease, the sounder, and therefore the more effective will be the methods we adopt for its prevention, relief, or cure.

It therefore becomes necessary to consider whether we have among us at this present time men skilled in scientific inquiry. That there are many in our profession capable of prosecuting difficult scientific investigations, must be clear to every one who will be at the trouble of glancing at the transactions of the Royal, Medico-Chirurgical, Chemical, Microscopical, and other learned societies. It is well known that many distinguished chemists and microscopists have been active medical practi-

tioners, while one of the most distinguished physicists of modern times (Mayer) is a medical practitioner. But there can be no doubt that the number of men in our profession capable of undertaking and actually engaged in scientific research is much less than it ought to be, and than it would be in this country, if the public understood the great value of scientific medical work. This is, perhaps, to be attributed partly to the idea that scientific work unfits a man for a practical calling, and partly to the impression that the public has no confidence in any but *practical*, as distinguished from *scientific*, doctors. But it is scarcely necessary to observe—for every reasonable person must now be convinced of this—that a thorough scientific training and thorough scientific work cannot but make a man a more thoughtful, and necessarily a more careful and judicious practitioner; while it is obvious that such a person must have advantages in investigating a difficult or doubtful case over one who has not so studied.

Of all departments of scientific inquiry, those bearing upon the study of the changes taking place in and upon the nature of disease, may be most conveniently and successfully prosecuted by medical practitioners. Some of us in active practice have had laboratories and work-rooms in our houses, and it is obviously possible for a man even in very large practice, whose time is much occupied, to employ assistants under his immediate direction who would assist him in detailed investigation. I am sure that if greater facilities were afforded for the scientific investigation of disease, and the work respected as it deserves, there would be no lack of workers. It is, indeed, surprising, that those connected with our great hospitals have not, long ago, taken steps to facilitate the advancement of medicine, instead of devoting their energies and means solely and entirely to the relief of present suffering. There is as yet, I believe, only one hospital in London in which there are efficient means for conducting scientific inquiries into the nature of disease, and I do not believe there is one, the managers of which would allow a very moderate sum, say £300, to be set apart for working expenses.

Not a few benevolent persons will perhaps think that the scientific investigation of disease means, in plain English, performing scientific and necessarily unjustifiable experiments upon the sick poor. Nothing of the kind, however, has been thought of by those who are so anxious for the prosecution of scientific medicine. The minute investigation of the secretions, chemical and microscopical; the careful study by the aid of delicate instruments of the state of the pulse and the breath; the analysis of the air breathed by the sick, as well as that of the surrounding atmosphere; the minute examination of diseased organs after death, are points upon which exact information would probably enable us to draw very important conclusions with reference to the nature of many serious diseases, while such inquiries would in no way affect the patient. All

that is required to carry out such work is well-arranged laboratories and work-rooms in our public hospitals, and qualified officers to do the work. The expense would probably not exceed £500 a year. What the practical results would be if such work were carried out in six or eight of our magnificent hospitals, it is of course impossible to say, but upon very many grounds we are justified in concluding that the gain to medicine, and indirectly to the public, would be considerable.

The objections previously urged to attempts to organise a large staff of scientific observers for the purpose of prosecuting a special inquiry, apply with equal force to practitioners, as to purely scientific men. It therefore follows that, practically, we are compelled to depend upon the efforts and energies of individuals. How then can individual energy be employed most advantageously? It appears to me that, upon the whole, the plan most likely to succeed is an extension of that system which has been for many years in operation in the Royal Society and is adopted by the British Association. It seems probable that if the Government would make grants to those who were engaged in scientific researches upon questions of such great public importance as the investigation of special forms of fever in man and animals, cholera, &c., great encouragement would be afforded. The number of workers would soon multiply, and it could scarcely happen but that many new facts would be demonstrated, and, from time to time, discoveries, of which the country might feel justly proud, would be made. Grants would not only enable men to enter upon expensive inquiries which they could not otherwise undertake, but would excite a taste for purely scientific inquiry in younger men. I dare say that of a great many grants made, several would be unproductive of results, but it seems to me that if, out of twenty men set to work, only one produced anything of value, the system would have been proved to have been successful. In the case of guns and ships how very much we are compelled to spend upon experiments which turn out complete failures.

I am only pleading for the expenditure of a very small annual sum upon the scientific investigation of diseases of the utmost consequence to the public—say £5,000 in grants, or from £50 to £200 to different individuals—and I think that if this sum, or as much of it as might be applied for, were voted annually, very satisfactory results would be apparent in the course of a few years. Many objections may possibly be urged against this plan, and objections may be raised to any proposal that might be made, but I think that a careful perusal of the reports of the British Association for the Advancement of Science will satisfy any one that the system of grants upon the whole has worked well, and especially in the case of the Kew Observatory, which has been the largest recipient for many years past.

But the same object might be attained in another way, and im-

mediately, if the public will give their sanction and support. We have many ancient corporations with vast funds at their disposal, which are, in some instances, devoted to a purpose which is not altogether approved. The work I am advocating might be efficiently promoted and be paid for by a fraction of the sum spent in unnecessary feasting. It would be impossible to propose a more useful way of applying a small portion of the income, for not only might great benefit result to the poor now living, but the public would reap advantage, and not only now, but in the future, and a scientific report might excite animated conversation at many a social entertainment. Laboratories and work-rooms for the microscope ought to be attached to all our hospitals, and sufficiently extensive, not only for the prosecution of original investigations, but also for teaching. In no other manner can the advance of medical science be so surely promoted. In this country it is true that much—few are aware how very much—has been achieved by the individual energy of many members of the medical profession in their own private work-rooms, but in such a city as London, if the suggestions offered were acted upon, more advanced scientific work might be carried out with facility in a year than is now performed, under great difficulties, in ten or twenty.

Every hospital might employ a small portion of its income in thus advancing our knowledge of disease, and surely the subscribers would not object to the appropriation of money to such a purpose. The benefit accruing from such investigations is felt, not only by the poor, but by the rich; but because such investigations are not often of much advantage to the rich of the present time, philanthropists look coldly upon them, and even oppose every kind of earnest thoughtful investigation. But surely it would be right if rich bodies, like Guy's, Saint Thomas's, and Bartholomew's took the lead in this matter. One would think that £1,000 of their large incomes might be spent very advantageously in scientific work, but I fear it will be difficult indeed to convince the authorities who have command of the purse. In some hospitals, such is the feeling of those who administer the funds that the question has been raised whether the charity should provide, for physicians and surgeons—who freely give their services to the institution—thermometers, microscopes, and many other scientific instruments, which are as necessary for the investigation and proper management of some cases as are bandages, surgical instruments, and other appliances for the treatment of others.

The reader must not infer that I am advocating scientific investigation, and the establishment of laboratories and work-rooms as a theorist, or only because such institutions have been established abroad. I have had considerable experience in the matter, for, upwards of twenty years ago, I fitted up an institution of the kind, for chemical and microscopical

work, in connection with medicine and pathology, close to the hospital in which I worked. Here, for several years, I taught privately, held classes, and gave lectures on the use of the microscope and on the chemical and microscopical investigation of the tissues and fluids of the body. In fact this book, which was based upon one of my courses of lectures given in 1853, would probably not have been written had I not commenced early in life to prosecute scientific work, in connection with clinical investigation.

The persons to conduct advanced scientific inquiries, in connection with medicine, are undoubtedly the young physicians and surgeons attached to our medical schools and hospitals. Were but a little encouragement afforded, I am sure that many who are eminently fitted for such work would willingly study, here and abroad, so as to perfect themselves in the branch of investigation they desired to pursue, and thus become highly skilled original inquirers. And when I say, if "a little encouragement were afforded," I mean, if a place in which they could work was found for them, and an income just sufficient to provide the necessaries of existence—say, £100 a year. Would not many a talented young physician and surgeon be better employed in spending part of his time thus than in devoting himself for fifteen or twenty years to seeing out-patients?—but, in fact, he might spend part of his time in scientific work and part in treating the sick. At this very time there are, probably, upwards of two hundred highly educated men in London, many with a taste for scientific work, and with intellects eminently suited to engage in it, who day after day are condemned to pass several hours in the routine work of seeing and prescribing for from fifty to a hundred patients per hour.

It is much to be regretted that a few distinguished surgeons, who unfortunately are able to influence the opinion of non-medical governors of hospitals, have expressed themselves very strongly against scientific investigation, and especially microscopic inquiries in connection with practical medicine and surgery. No doubt, by acting thus, students may be prejudiced against science, and persuaded to remain ignorant of many things they ought to know, but the teacher who encourages idleness or contempt for real work and knowledge is not likely to receive the thanks of his pupils in after life. So curiously intense is the feeling in the minds of some of the senior and most influential members of the profession against the scientific investigation of disease that there is, at present, little hope of convincing the governors of our great public medical charities that money should be spent in the manner proposed. The quiet determined opposition to the advance of scientific medical knowledge in the profession itself is very curious, but much to be deplored. Not very long since, a most distinguished French surgeon expressed a very strong opinion against microscopical investigation, and his views

would, I fear, be endorsed by more than one British surgeon, who has no knowledge whatever of any kind of microscopical inquiry. Unprejudiced persons ought not to be influenced by the microscopical condemnation of surgical authorities who have never studied, and have determined not to learn anything in connection with the subject, especially as many facts recently made out are of the greatest importance to practical surgery, and ought to be known to all surgeons. One wonders that professional authorities, who are opposed to particular methods of inquiry into nature's secrets, do not authoritatively forbid them to be employed under any circumstances, and enact that any candidate for surgical diplomas who shows a knowledge of the microscope should be rejected, and that, at least in surgery, the microscope shall not upon any account be employed.

The strongest opposition, however, can only retard progress. Happily, there is now no power in Europe that can prevent those who desire so to do from learning and working, although it is to be regretted that, among those who call themselves *liberal*, a very decided longing to exercise tyranny is occasionally exhibited, and the duty of warning the ignorant against relying upon the judgment of a brother doctor who was known to use a microscope has been too conscientiously discharged. However, as every year adds to our knowledge many new scientific facts, bearing in the most important manner upon our views of the changes which occur during the healing of wounds, it is obvious that hostile doctrines may be left to die out, and the hard words that have been used may be forgotten. Opposition to scientific investigation, in connection with any department of medicine and surgery, will soon be considered ridiculous by thoughtful persons, and, although young students may be perhaps not unwillingly misled and deceived for a time, they, too, will soon discover the truth and use their reason. Many, after having passed their examination and entered the profession, find it necessary to study matters which they ought to have learnt during the period of their pupilage, but which they were told were useless and unpractical.

It must be obvious to any intelligent person that, by careful microscopical inquiry and chemical investigation alone, can we hope to gain definite information concerning some of the most important phenomena of disease. By pursuing such investigations great benefit will unquestionably be conferred upon the poor, and it is doubtful whether the same amount of money laid out in any other manner would be productive of greater advantage to mankind.

I have been advocating the prosecution of certain lines of investigation, which will to a certainty be followed out, whether or not they be encouraged by the State, by societies, or by wealthy individuals. Some, no doubt, will agree with me in thinking that the private energy and

enterprise of individual scientific workers who are often as poor as they are intelligent, devoted, and skilful, ought to be occasionally supplemented and assisted; while others may think it right to leave the investigators alone, until the results obtained are so very decided and so valuable as to command public attention. But even in this case there is no hope of direct remuneration or reward, for as soon as the work is completed, it at once becomes public property. The results of scientific research cannot be protected by patents and have no money value whatever, though they may be the means of saving the lives of thousands and benefit civilized society. Concerning the nature of disease, it is certain that very much more than is yet known will be discovered—nay, more than seems to us likely at this present time ever to be discovered. The only question for the public to determine is whether such scientific investigations shall be urged on at a rate commensurate with the enormous means and power of work at disposal in this country, or left entirely to the very few who can pursue science unaided. We may perform our part in the great work, or we may permit the poorer Continental States to be the sole employers and supporters of purely scientific labour, of which it is no doubt true we shall share the results though they will, of course, have, as they deserve, all the credit of encouraging the progressive work.

Most scientific men, at any rate during the earlier and more active part of their working days are poor, and those who work the hardest and most skilfully are often the poorest, and being the poorest have necessarily very little influence upon the opinions of statesmen who alone have the power to make laws, and influence to obtain a vote for money-grants for original scientific investigations.

It must, I fear, be admitted that the man who pursues unremunerative work, will, in these days, from that circumstance alone, be looked down upon by the more fortunate persons who gain money by the performance of less difficult operations. A poor scientific worker is too often regarded as an unpractical half mad enthusiast, or deemed a silly fellow, or mean drudge who is only able to make but a contemptible pittance by his work. For his labour will only command low wage and by many will not be considered to possess any money value at all. There never has been any demand for original scientific medical work. Were the scientific man to stand in the market place, no one would hire him. By commercial persons he will be accused of being a mere spender of money instead of being a producer of wealth. No one would pay for his most brilliant results the price of the paper on which they are recorded, however important they might ultimately prove to be to mankind. To frugal money-accumulating citizens, scientific investigation may seem to be unmeaning extravagant waste. To encourage men to spend their lives in scientific work is perhaps

considered on a par with fostering that very undesirable class of persons who consume the material resources of the state without giving anything tangible in return. Nevertheless, in spite of this and much more that might be urged to the same effect, and notwithstanding the discouragement of many influential people, and active opposition on the part of some, scientific work must be prosecuted, and he who has a real love for his profession and the necessary qualifications, will assert his independence and devote at least a part of his time to advancing some branch of scientific investigation which may be likely to promote the interests of medicine.

To my mind, however, the most depressing feature of the present tendency is the serious opposition to original work on the part of certain scientific men themselves. For years past there has been in England a very powerful and most active scientific party which has distinguished itself rather by its intense opposition to those who differed from it than by its own quiet prosecution of scientific investigation. It has done very much but it has discovered little. It has unfairly exalted in public estimation the work of foreigners that seemed in accordance with its acknowledged tendencies, and yet more unfairly disparaged the work of Englishmen that was in any way opposed to its extravagant pretensions. The "laws" it has succeeded in making people acknowledge would in any other country have destroyed original investigation and well nigh rendered impossible individual thought and action opposed to it. This party seems to believe that original enquirers, like varieties of pigeons, are to be made according to order. By stringent examinations it was proposed that every good intellect should be forced to run in the same rut. By extending these over many years it was supposed that all might be successfully drilled into uniformity, and thus a thoroughly well-trained scientific rank and file ensured. But in so far as these great objects have been attained, has individuality been crushed out; and, of those who most distinguished themselves in competitive examinations, far more have acted up to the ambition to achieve success in what paid best, than have devoted themselves to the quiet steady prosecution of original enquiry. But now that the evil has been discovered, an infallible remedy is of course at hand. Original investigators are in future to be trained according to order, and then, having been properly provided for, are to be confined in the special original-work pen, there to pass their existence in the secretion of new facts. That such things should be proposed is not perhaps very extraordinary in these days, but that sensible people should be prevailed upon to give effect to proposals so absurd, is unfortunate. That Mr. J. S. Mill should have to write as follows in the year 1873, is sufficiently significant:—"The abolition of the competitive examination for fellowships seems to me the reverse of an improvement. I quite understand that

the object of this proposal is to prevent the appointments from being obtained by cramming. But it is not beyond the capacity of the Universities to take sufficient security that success in the examinations shall not depend on cram; nor is it understood that the high honours at either Cambridge or Oxford are generally so obtained. On the other hand, I have the greatest distrust of all schemes for disposing of high and well-paid employments by a nominating body. Such bodies, having only a collective responsibility, are often even more addicted to abusing their patronage than high functionaries; the members are apt to job for one another, and vote for each others' *protégés*. And even without the supposition of jobbing, a body like that you have in view, composed indeed of scientific persons, but of persons whose position and reputation are already made, is not at all likely to look with favour on the striking out of new paths. *Experience shows that academies, whether of literature or of science, generally prefer inoffensive mediocrities to men of original genius.**"

* "Athenæum," November 1st, 1873. Letter to Mr. E. Ray Lankester, on being asked to join the "Association for the reorganisation of Academical Study."

PART I.

THE APPARATUS NECESSARY FOR THE EXAMINATION OF OBJECTS OF CLINICAL IMPORTANCE—OF THE PRACTICAL OPERATIONS REQUIRED FOR THEIR DEMONSTRATION—OF RECORDING THE APPEARANCES OBSERVED.

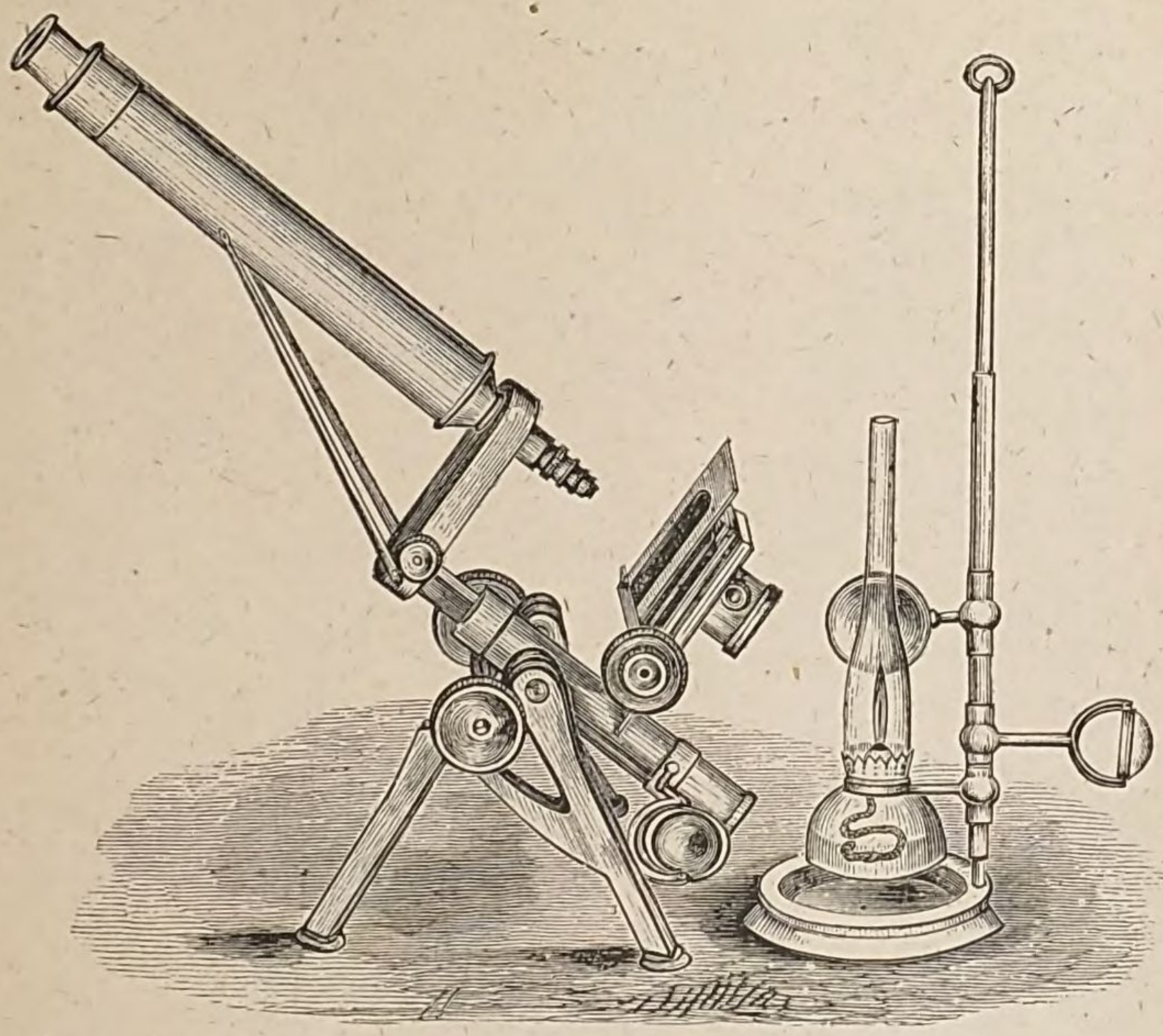
CHAPTER I.

Of the Apparatus necessary for Microscopical Investigation.—Method of submitting a portion of Tissue or other Object to Microscopical Examination.—Of the Medium in which Objects should be Examined. Great caution necessary in drawing inferences from Microscopical Appearances.—Of drawing Objects.—Camera Lucida.—Steel Disc.—Glass Reflector.—Of Drawing Objects which it is intended should be Engraved.—Of ascertaining the Magnifying Power of Object Glasses. —Of Measuring the Diameter of an Object.

In this work I shall endeavour to give information which is likely to be practically useful to the student of medicine and practitioner in the exercise of their duties in hospitals and in private practice. Many of the methods of investigation I shall describe will be found useful to those who prosecute microscopical research as a scientific pursuit, as well as to practitioners who may be called upon to undertake investigations as scientific witnesses in medico-legal cases or as officers of health.

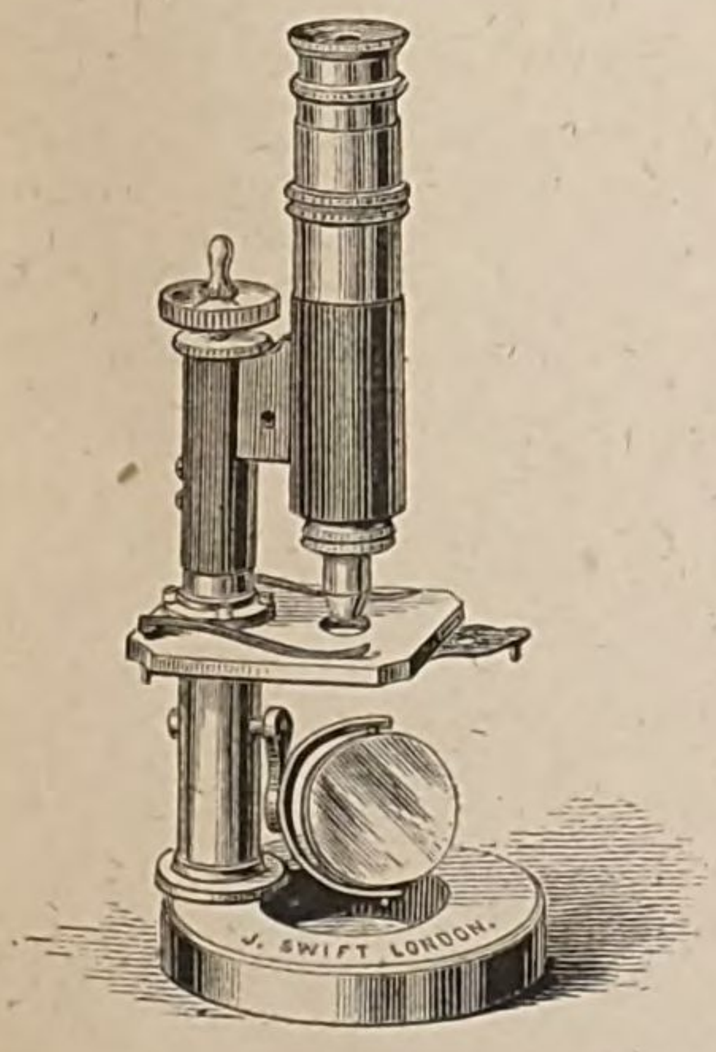
To medical practitioners a good knowledge of the use of the microscope becomes of greater importance year by year, and there are, it need scarcely be said, many cases, the nature of which cannot be surely determined without its aid. Moreover, practitioners are now-a-days expected to be able to report upon the characters of food and water, and to undertake many enquiries which cannot be prosecuted by any one who has not acquired a knowledge of the methods of conducting minute chemical and microscopical examination, and has not gained experience in the manipulation required in minute research. In order to make my work practically useful it will be necessary to refer to the different instruments and pieces of apparatus specially required, but I shall be as brief as possible, for most of the instruments and pieces of

Fig. 1.



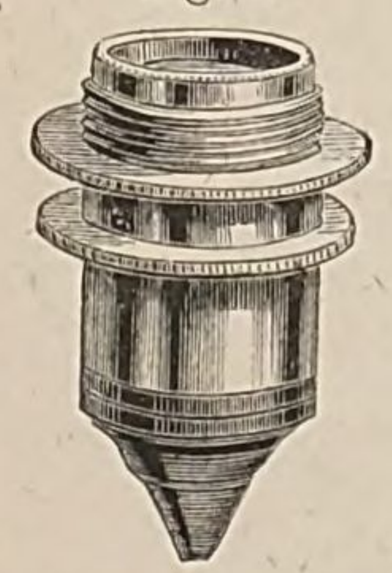
Position of microscope and lamp for viewing objects with high powers. The light passes at once to the condenser, instead of being reflected from the mirror. p. 26.

Fig. 2.



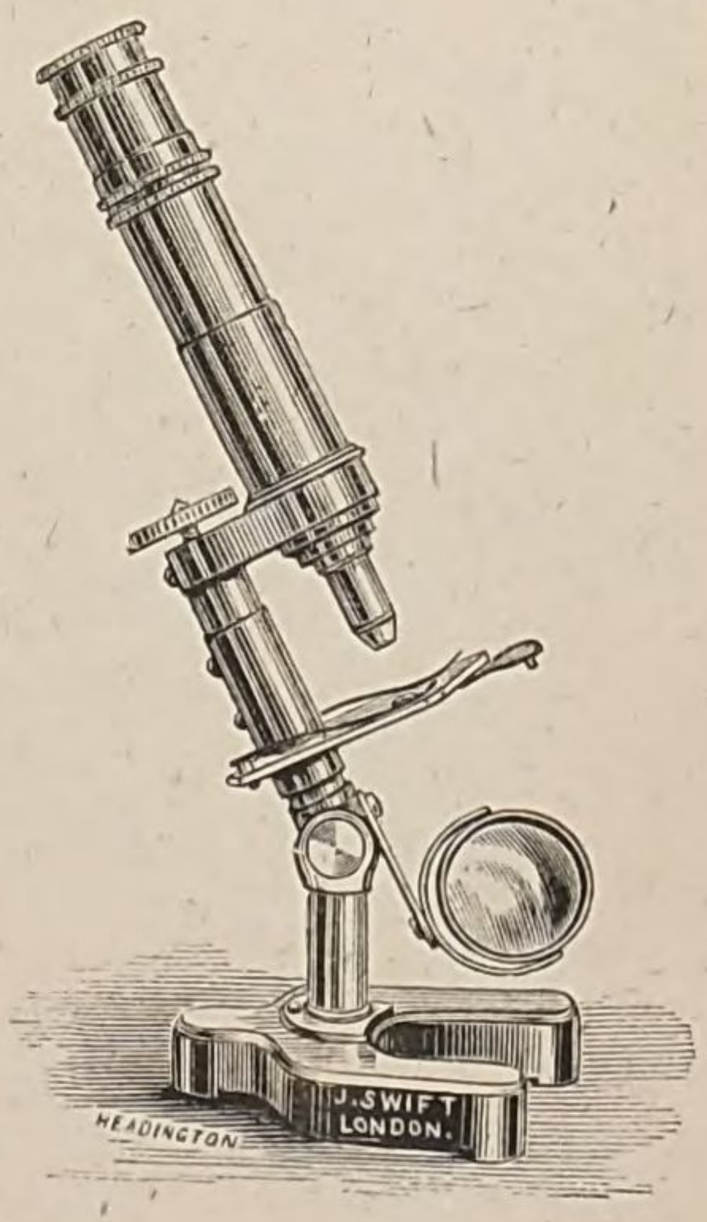
Small Microscope after Gundlach's Model, made by Mr. Swift. p. 22

Fig. 4.



Half-inch objective, recently made by Mr. Swift, made taper, to facilitate the use of the bull's-eye condenser.

Fig. 3.



The same instrument as Fig. 2 inclined.

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accessory apparatus have been described in "How to Work with the Microscope," where also will be found a more detailed account of instruments requisite for special microscopical enquiries other than medical. Frequent references will be made to the different sections of that book. For the sake of brevity the following simple form will be adopted:—H. to W., §.

Of the Microscope and Accessory Apparatus.

1. Microscope.—In the *simple microscope* the magnified image of the object passes at once to the eye of the observer. The *compound microscope* is the only one now used for microscopical research. In this instrument the object is magnified in the first instance by the *object glass*. This magnified image is further magnified by the *eye-piece*. The last image is of course inverted, which inconvenience may be obviated, if desired, by causing it to pass through another set of lenses termed the *erector*.—H. to W., § 3.

The *Student's Microscope* should have a large stage, firm tripod stand, coarse and fine adjustments, double mirror, and arrangement for inclining the body. With two powers and bull's-eye condenser a good instrument costs from five to ten guineas. Some forms of students' microscopes are referred to in "How to Work with the Microscope," §§ 15, 16, where the chief points to be considered in choosing a microscope are enumerated. Students' microscopes are now made by almost all microscope makers. To Mr. Salmon, however, is due the credit of being one of the first to make a well-arranged cheap microscope.

Some observers, especially Professor Hughes Bennett of Edinburgh, find great fault with English-made instruments, and highly praise the small German microscopes. Undoubtedly, many French and German instruments are very good and *some* English ones are bad, but it is as unjust as it is ridiculous to condemn English microscopes wholesale and call them bad names. The very best instruments I have ever seen have been made in England. I assure the reader that with the aid of a good English microscope, objects can be examined with very high powers and brought to a focus, the powers changed, and the specimen looked over in every part in less than half the time these operations can be conducted under a foreign instrument. Any one who has worked with powers above the eighth, and has discovered how to use a movable stage properly, will have been convinced of its advantages. Those who condemn this part of the microscope can have had little experience in prosecuting any of the departments of microscopical research in which very high powers are required.

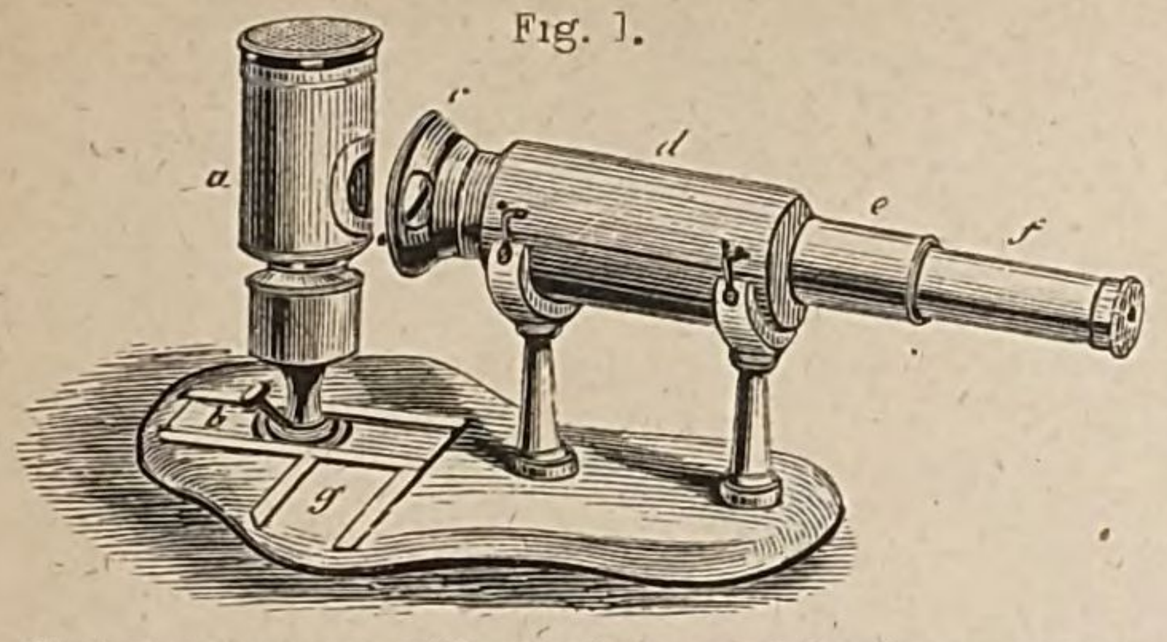
To examine tissues with a twenty-fifth, a good movable stage is almost indispensable, and as much work may be done with its aid in an hour as can be performed without it in half a day. Still there are

gentlemen who speak with stern authority, who tell us that such high powers are not required and that nothing is to be learnt by them. All I shall say on this head in this place, is, that I trust no one who desires to enter upon real work will allow himself to be influenced by such vague prejudiced statements, or be misled by views upon highly important anatomical questions however confidently stated, which are based upon observations made with powers magnifying less than five hundred diameters, upon specimens prepared by immersion in Canada balsam or some highly refracting resin, or immersed in water, vitreous humour, or white of egg. It need, however, scarcely be remarked, that a student beginning work has much to learn before he requires any of those refinements absolutely necessary for one who intends to pursue original research. To the young student a cheap foreign microscope will be useful enough. Hartnach's students' microscopes are unquestionably very good. Gundlach's microscope is very efficient and one of the cheapest that the student can obtain. Mr. Swift is now making instruments according to this foreign model, pl. I, figs. 2 and 3, which, with one-sixth of an inch object-glass, packed in a case, costs only £3 17*s.* 0*d.* Still cheaper arrangements have been suggested. Some expense is saved by having the body of the microscope screwed into the box in which it is kept, but such an instrument is not so steady as that provided with a heavy metallic foot. The above and many other forms, some of which are figured and described in H. to W., are suited for all ordinary medical microscopical enquiries, but if the student should determine to prosecute microscopic investigation thoroughly, and to use high powers, I cannot do better than advise him to purchase one of the best stands, as that of Messrs. Powell and Lealand, fig. 1, pl. I, with an inch and quarter of an inch object-glasses, and subsequently add the higher powers and accessory apparatus he may require.

Of the Binocular Microscope.—The binocular arrangement cannot be said to be required for medical microscopical work. For the pursuit of some branches of the subject it would be advantageous, but for general work the single body is, in my opinion, to be preferred. Any one desirous of having a binocular, can have the double tube fitted to his instrument if it be a good one, and in this case the double tube is so adapted, that it can be removed at pleasure.

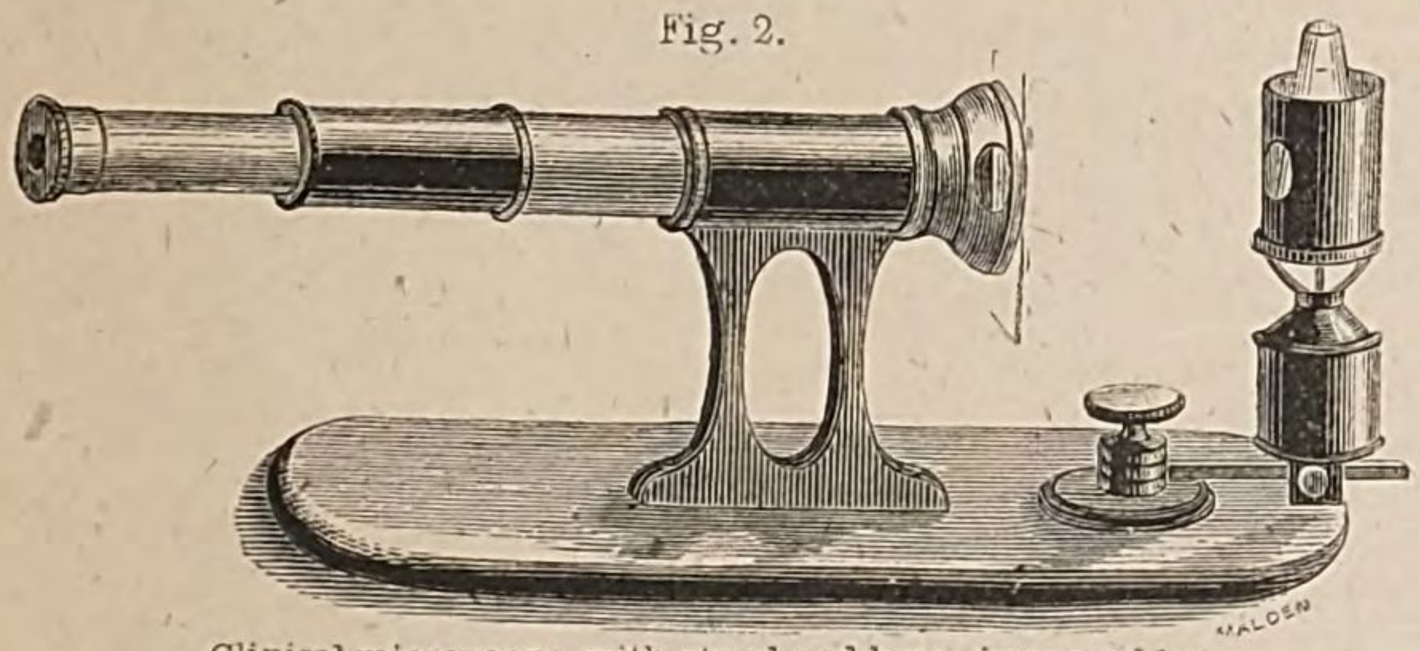
Of the new Binocular Arrangement adapted for the Highest Magnifying Powers.—Messrs. Powell and Lealand have recently succeeded in devising a plan by which a binocular arrangement is adapted to the highest powers. The ordinary binocular now in use is suitable only for the examination of objects by powers magnifying less than 200 diameters, but the new one can be used with the $\frac{1}{50}$. By the prisms represented in section in pl. III, fig. 4, it will be observed that of the

Fig. 1.



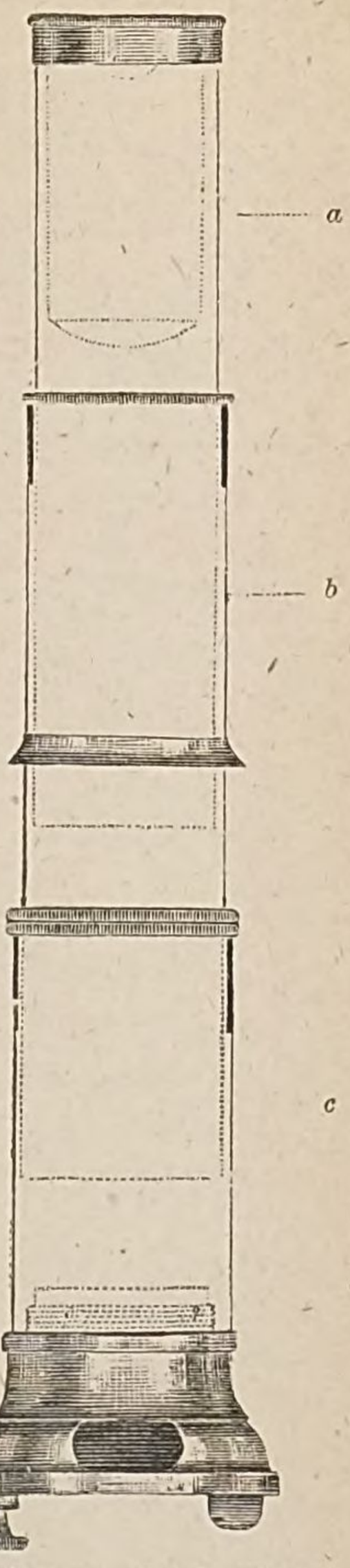
Clinical microscope with stand, arranged for class purposes, with lamp. § 2. p. 23.

Fig. 2.



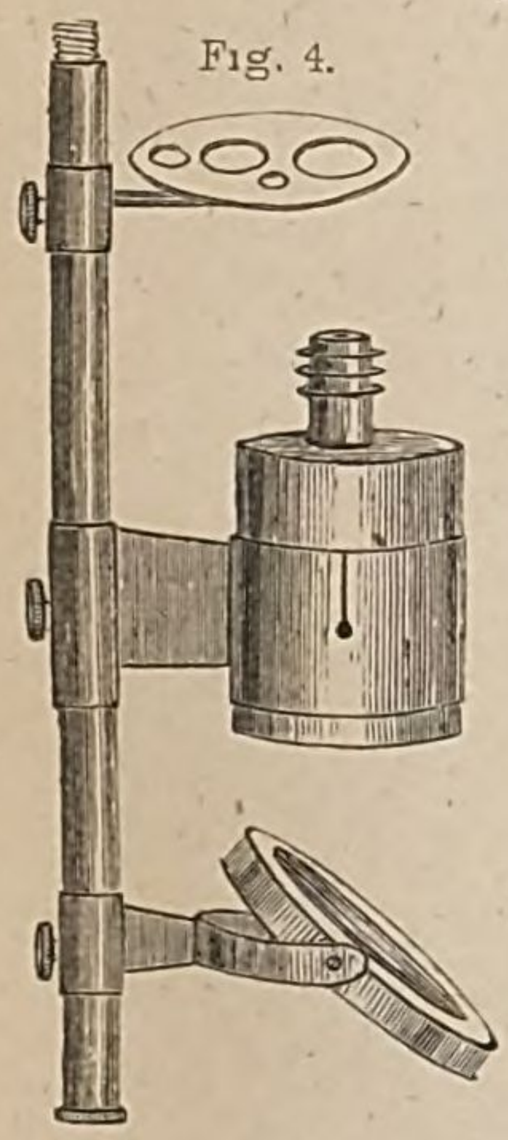
Clinical microscope, with stand and lamp, improved by Mr Highley. § 2. p 23.

Fig. 3.



Pocket or clinical microscope, half the real size *a*, tube with eye-piece. *b*, tube carrying object glass. *c*, tube in which the last slides, with stage. *e*, clamp for fixing preparation. § 2. p. 23

Fig. 4.

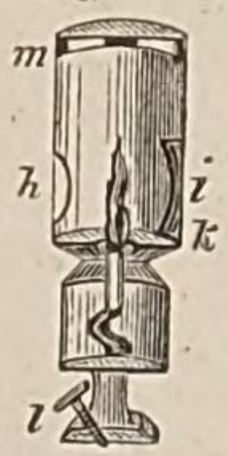


Arrangement for adapting diaphragm mirror and condenser to 'clinical microscope.' § 2.

Fig. 5.

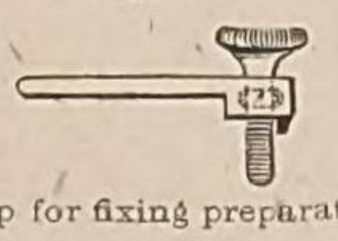
Mirror for clinical microscope. § 2

Fig. 6.



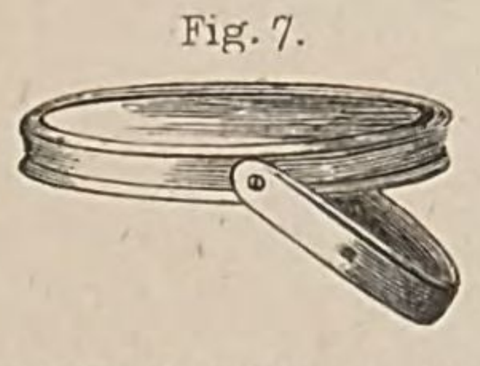
Lamp. *h*, aperture with mica. *k*, diaphragm, *l*, screw for fixing lamp § 2.

Fig. 6*.



Clamp for fixing preparation

Fig. 7.

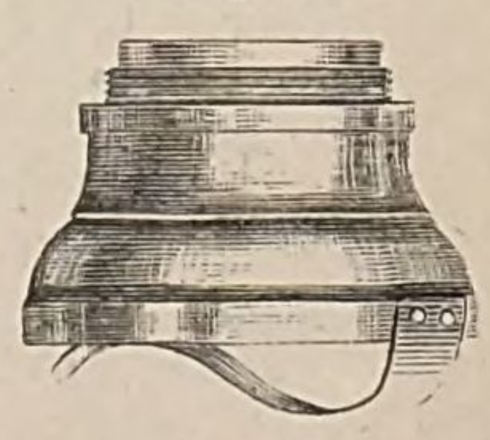


Mirror employed for examining objects by transmitted light. § 2.

Fig. 8.

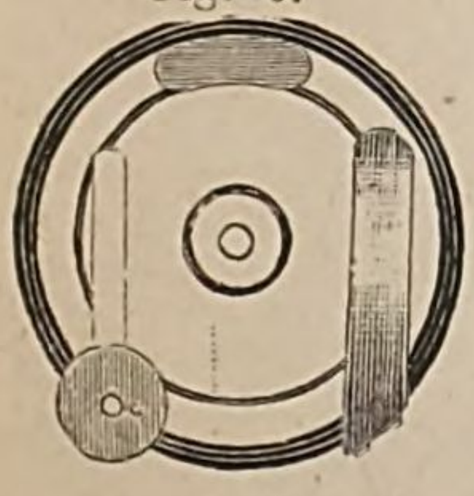
Sectional view of cell.

Fig. 9.



The stage, side view, showing position of the spring. § 2.

Fig. 10.



The stage (fig. 9) seen from below.

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total number of rays which have passed through the object glass, the greater part are transmitted through the prism B and the straight tube of the microscope, but some suffer reflexion from its lower surface, and are received upon the reflecting surface E of the prism C in an oblique direction as shown by the dotted lines, and after emerging from the surface, enter the diagonal tube of the microscope. I have examined many objects by the arrangement recently adopted by Messrs. Powell and Lealand, and find that it works exceedingly well in practice, and I can strongly recommend it to those who propose to work with the highest powers yet constructed.

2. The Clinical, Pocket, Travelling, and Class Microscope—Is an instrument devised by me some years since, which I have found very useful for general observation in the fields, and also for medical work and for class demonstration. This form of microscope is composed of draw-tubes like a telescope, of which there are three, tube *a* (fig. 3, pl. II) carries the eye-piece, is four-and-a-half inches in length, and slides in tube *b*, which is of the same length, but only slides up to its centre in the outer tube, *c*. Tube *b* carries the object-glass. The tube *b* can be fixed by the aid of a screw-ring, *d*, at any height, according to the focal length of the object-glass. This arrangement prevents the risk of the object-glass being driven through the preparation while being focussed. A screw clamp is attached to the lower part of the body for fixing the preparation in any particular position. There is also an aperture for admitting light to opaque objects. The preparation is held in close contact with the flat surface at the end of the microscope by pressure of a spring (fig. 9), which allows the requisite movements to be made with the hand.

That part of the object which it is desired to examine can be easily placed opposite the object-glass if the instrument be inverted. Next, the focus is obtained by a screwing movement of the tube *b*; and if it be desired to examine any other parts of the object, it may be easily effected by moving the slide with one hand while the instrument is firmly grasped by the other. Delicate focussing is effected by drawing the tube *a* up and down, a movement which alters the distance between the eye-piece and the object-glass.

Any object-glass may be used with this instrument. I have adapted various powers, from a *three inch*, magnifying *fifteen diameters*, to a *twelfth*, magnifying *seven hundred diameters*, and I feel sure that even higher powers may be used.

For transparent objects, ordinary daylight or the direct light of a lamp may be used. For opaque objects and for ordinary reflected light examinations, sufficient illumination is obtained from an ordinary wax candle or small lamp placed at a short distance from the aperture just above the object.

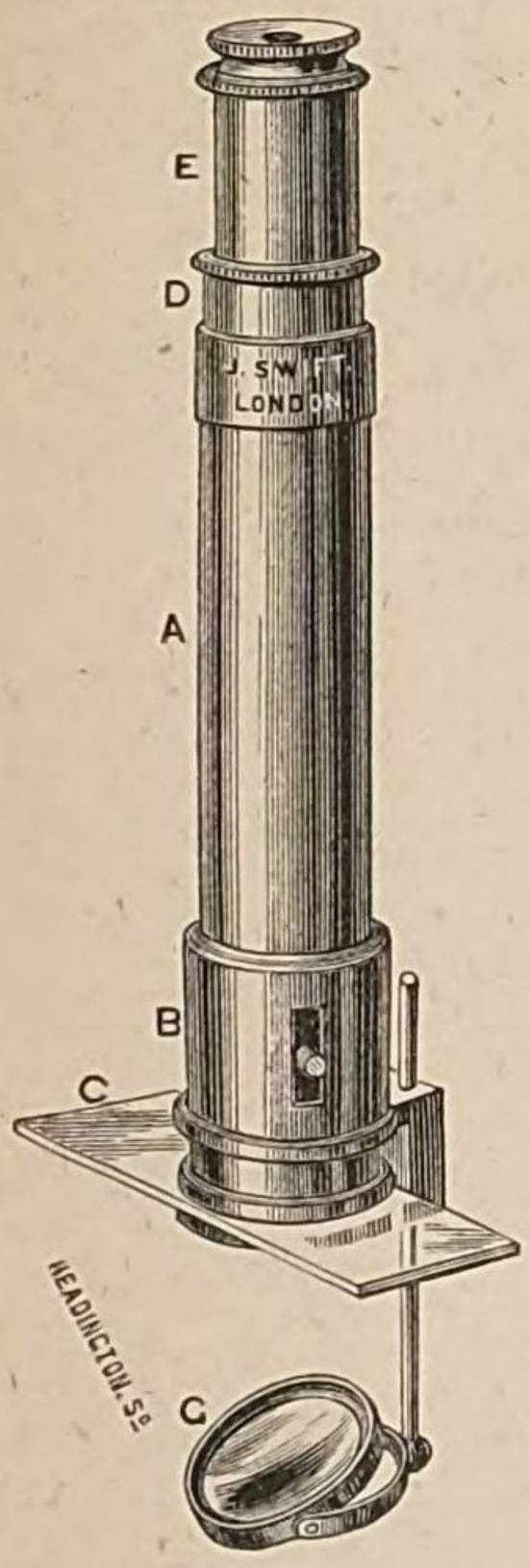
I have used this little microscope in clinical teaching. Various urinary deposits, specimens of sputum, &c., may be examined by the patient's bedside, and their characters demonstrated to the class, and it is most convenient for all ordinary microscopical work in the wards. It may be used either with or without its stand. The arrangement of the stand, with its lamp, mirror, diaphragm, condenser, &c., will be at once understood by reference to figs. 1, 2, 4, pl. II. These microscopes can be purchased for twenty-five shillings, without powers ; and with the stands they will probably cost not more than three pounds each. (See also H. to W., § 21.)

The clinical microscope has been modified and improved upon by many different makers. Very convenient forms have been introduced by Mr. Baker, Holborn, and Mr. Moginie, 35, Queen Square, and another by Mr. Hawksley, 4, Blenheim Street, Bond Street. Still more recently, a new form, arranged by Mr. Swift, University Street, is used as a clinical and seaside microscope, and with two object-glasses costs £4 10*s*. 0*d*.

Dr. Guy's Arrangement.—My colleague, Dr. Guy, has devised an ingenious form of hand microscope, which is now made by Mr. How, of Foster Lane, and is known as the "Illuminator Hand Microscope." This is well adapted for examination with low powers, and for showing popular objects in schools and to classes, but it is not adapted for the general work of the medical practitioner or student of physiology. My friend, in praising the merits of the class microscope he has constructed, has, I think, been unduly hard upon the complexities of other instruments, while he has not given due weight to the fact, that any microscope, to be of real service to the student and practitioner, must work well with a magnifying power of at least 200 diameters. In his paper he does not so much as even mention the highest amplifying power that can be adapted to the hand microscope he describes.—(Journal of the Quekett Club, No. 20, October, 1872, page 65.) In comparing and contrasting one another's instruments, we may mislead our readers, and unintentionally damage the reputation of our friends, if we are not careful in defining exactly the points considered excellencies or deficiencies in the case of particular instruments.

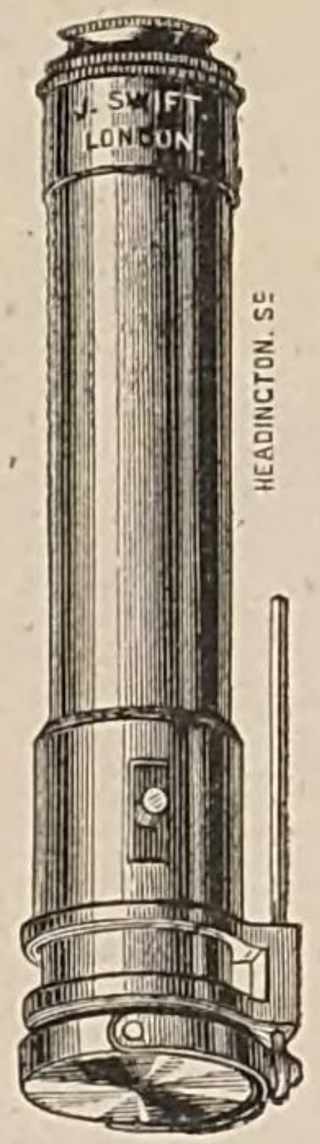
Pocket Microscope.—Professor Brown, of the Veterinary Department of the Privy Council, has lately introduced a very valuable modification of the clinical pocket microscope which occupies far less space than the one I have described. This beautiful little instrument can be used for examining objects under the highest powers, and can be carried in the waistcoat pocket. It is figured in plate III, figs. 1, 2, 3, two-thirds the actual size. This little microscope is four inches long by one inch in diameter in its widest part. The general arrangement of the instrument will be understood by the figures, but it has been fully described

Fig. 1.



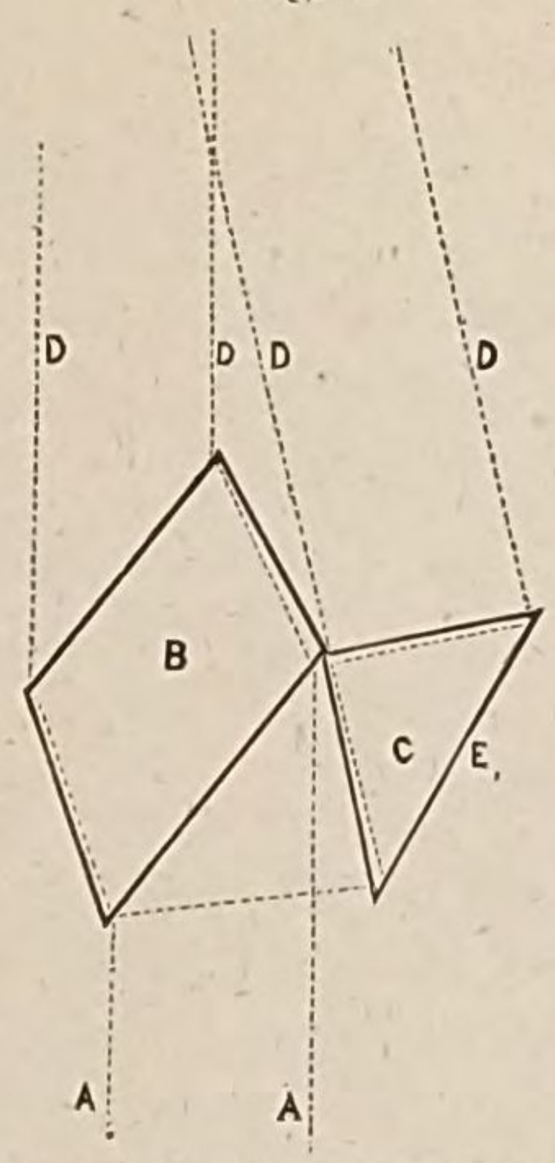
The Microscope (Fig. 2) arranged ready for use. p. 24.

Fig. 2.



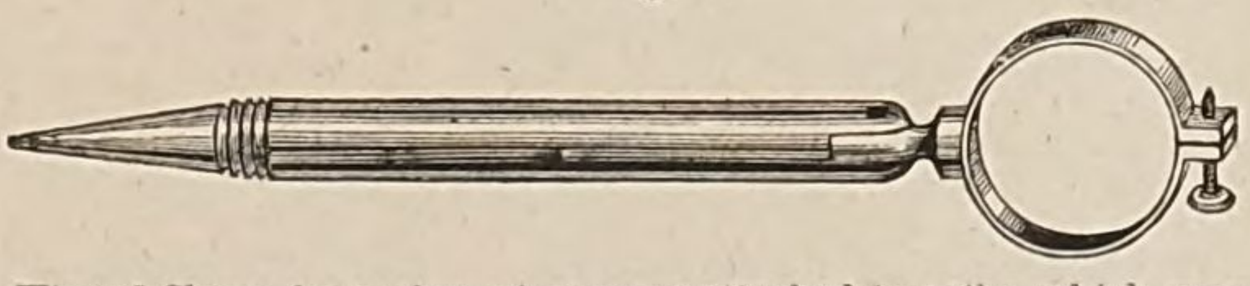
Prof. Brown's new Pocket Microscope, $\frac{2}{3}$ real size. p. 24.

Fig. 4.



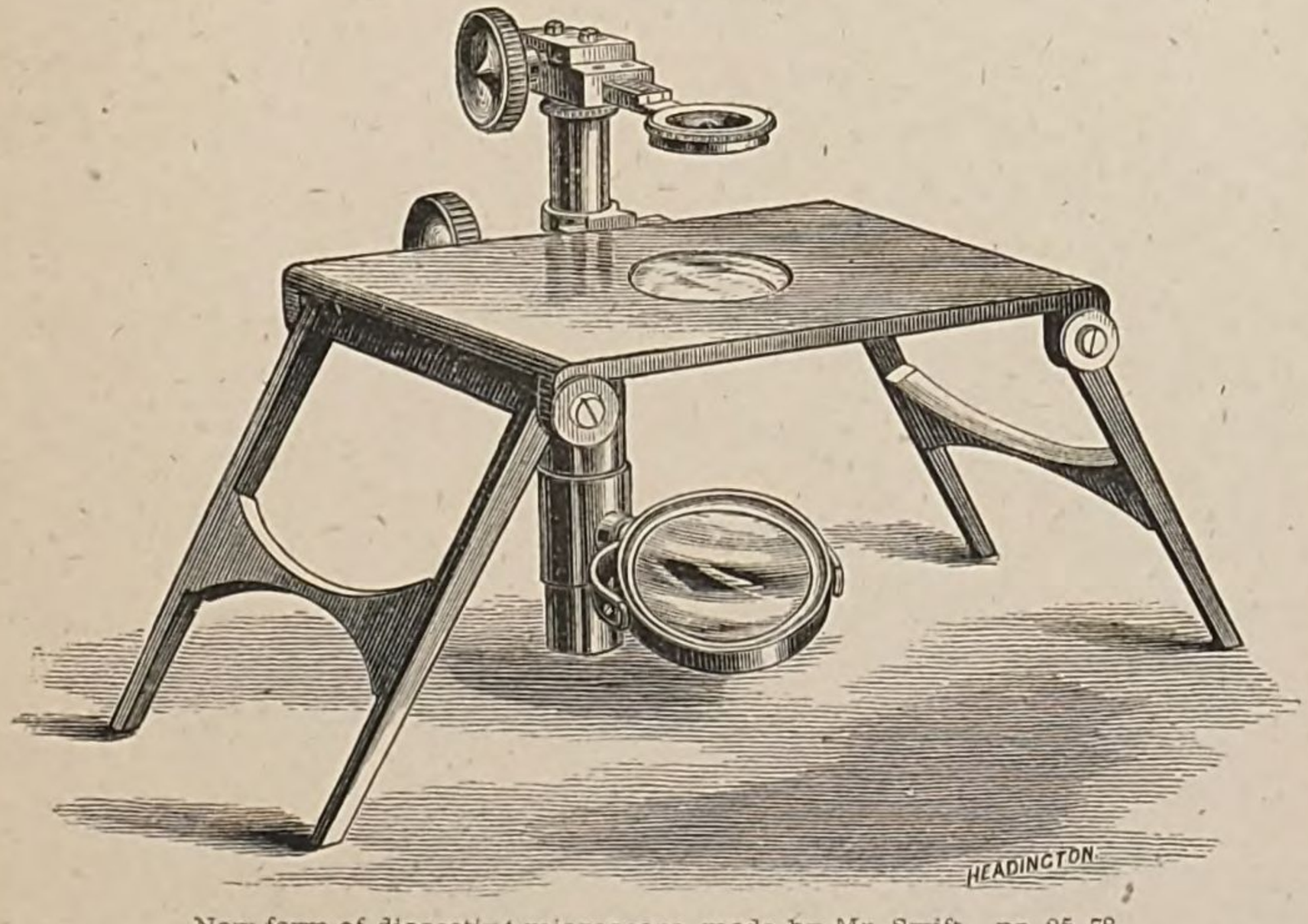
Powell & Lealand's binocular arrangement for high powers. AA, rays of light proceeding from object glass, B, parallel piece of glass. C, triangular prism DDDD, emergent rays. p. 22.

Fig. 3.



Hinged Clamp for pocket microscope attached to spike, which may be forced into a gate post.

Fig. 5.



New form of dissecting microscope, made by Mr. Swift. pp. 25, 78.

[To face page 24.

by Professor Brown in the "Veterinarian" for November, 1872, and it is made by Mr. Swift, University Street, Tottenham Court Road. The instrument, with a quarter, costs about three guineas. I believe this microscope will be found of the greatest use by members of the profession. A very little practice will enable any one to use it without difficulty. Professor Brown employs it in Veterinary work. He can use it in the open air, and can examine secretions, and the blood of animals in the sheds in which they stand, and under powers magnifying one thousand diameters. Condensers, polarizing apparatus, and other appliances can be added to the instrument without difficulty, if required. The glass slides, thin glass covers, pipettes, needles, &c., can be packed in the same little case with the microscope.

Dissecting microscopes are very useful in some medico-microscopical enquiries, but are not necessary for ordinary clinical work. Professor Lawson introduced some improvements. His instrument is represented in "How to Work with the Microscope." A good form of Quekett's dissecting microscope has lately been further improved by Mr. Swift. (See plate III, fig. 5.)

3. Eye-pieces—Negative and positive.—The eye-piece ordinarily used is the *negative* or *Hughenian* eye-piece. It consists of two plano-convex glasses, the *flat* surfaces of which are directed upwards. The glass nearest the eye is the *eye-glass*, and the one at the greater distance the *field-glass*. In the *positive eye-piece* the convex surfaces of the glasses are directed towards each other.

4. Object-glasses.—1. *The inch*, magnifying from 30 *to* 40 *diameters*, the glasses of which can be removed one by one, so that lower powers can be obtained. 2. *The quarter* of an inch, magnifying about 200 *diameters*. These glasses should *define well*, the field should be *perfectly flat* and free from *coloured fringes*, and they should admit a sufficient amount of light. The object-lenses used in the best instruments are of English manufacture, but some of those furnished with the cheap microscopes are made on the continent, and although much less expensive, the defining power of many of them is very good, insomuch that they are practically useful for all ordinary work. A good English quarter cannot be purchased for less than five pounds, but these foreign objectives can be obtained for from ten to thirty shillings. The higher object-glasses are useless without considerable practice. They are the *twelfth*, *sixteenth*, *twenty-fifth*, and *fiftieth*. The first of these magnifies about 700 diameters, the last nearly 3,000. These high objectives, now made by Messrs. Powell and Lealand, are most valuable glasses, but the student is recommended not to attempt to use them until he is perfectly familiar with the management of the lower powers and has had great experience in manipulation and the preparation of objects.

Immersion lenses possess many advantages, and are much cheaper than the old form. Some of Hartnach's and Gundlach's are excellent for the very low price charged.

Very good lenses of moderately high magnifying power are now made by several English makers besides the old well-known firms. The powers of Mr. Collins, Mr. Crouch, Mr. Baker, Mr. Moginie, Mr. Hawksley, are all good. I have lately seen a high power recently made by Mr. Swift, of University Street, Tottenham Court Road. One of his immersion sixteenths, which I possess, defines extremely well, and is in all respects a highly satisfactory glass.

5. The Diaphragm—Is a circular plate with holes in it of different sizes. By it the circumferential rays of light reflected from the mirror may be cut off.—H. to W., § 13.

6. The Bull's-eye Condenser.—This instrument is required for condensing the light on the object in the examination of opaque preparations, and for dissecting under the influence of a strong light. —H. to W., § 25.

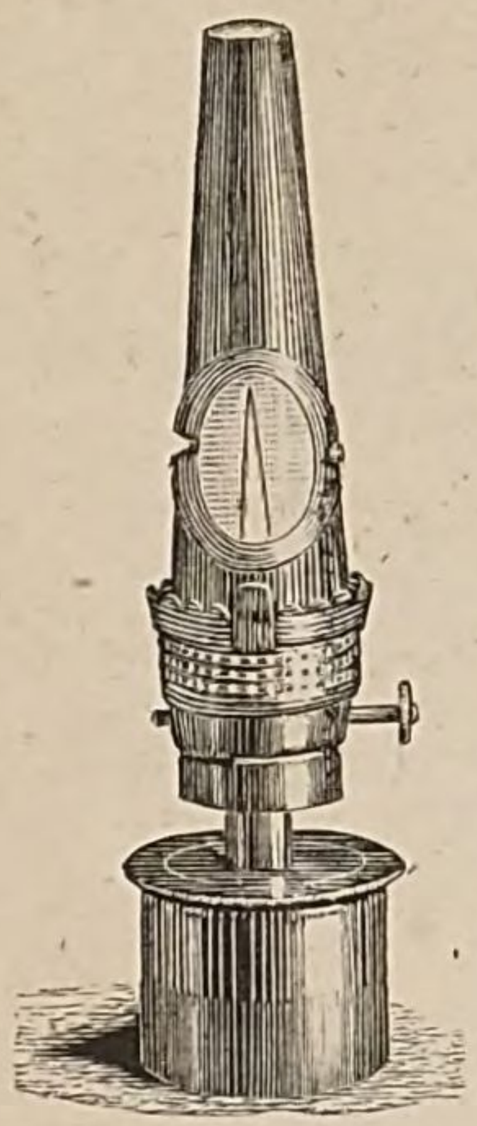
7. Achromatic Condenser.—Employed in the examination of objects by transmitted light.—H. to W., § 32. The most useful form of condenser is an inverted deep eye-piece, the smaller glass being covered with a thin blackened cover, having an aperture in the centre not more than the tenth of an inch in diameter. See pl. I, fig. 1.

An excellent universal condenser has lately been brought out by Mr. Swift, which combines the advantages of several separate instruments. It costs about ten pounds, but it is not required by those who are engaged in medical microscopical work only.

8. Lamps for Artificial Illumination.—A small French moderator forms an excellent lamp for microscopical work. The German lamps lately introduced by Mr. Pillischer give an excellent light, and can be easily arranged at any desired height. To microscopists provided with gas, I recommend the Argand gas lamp designed by Mr. Highley, fig. 4, pl. IV.

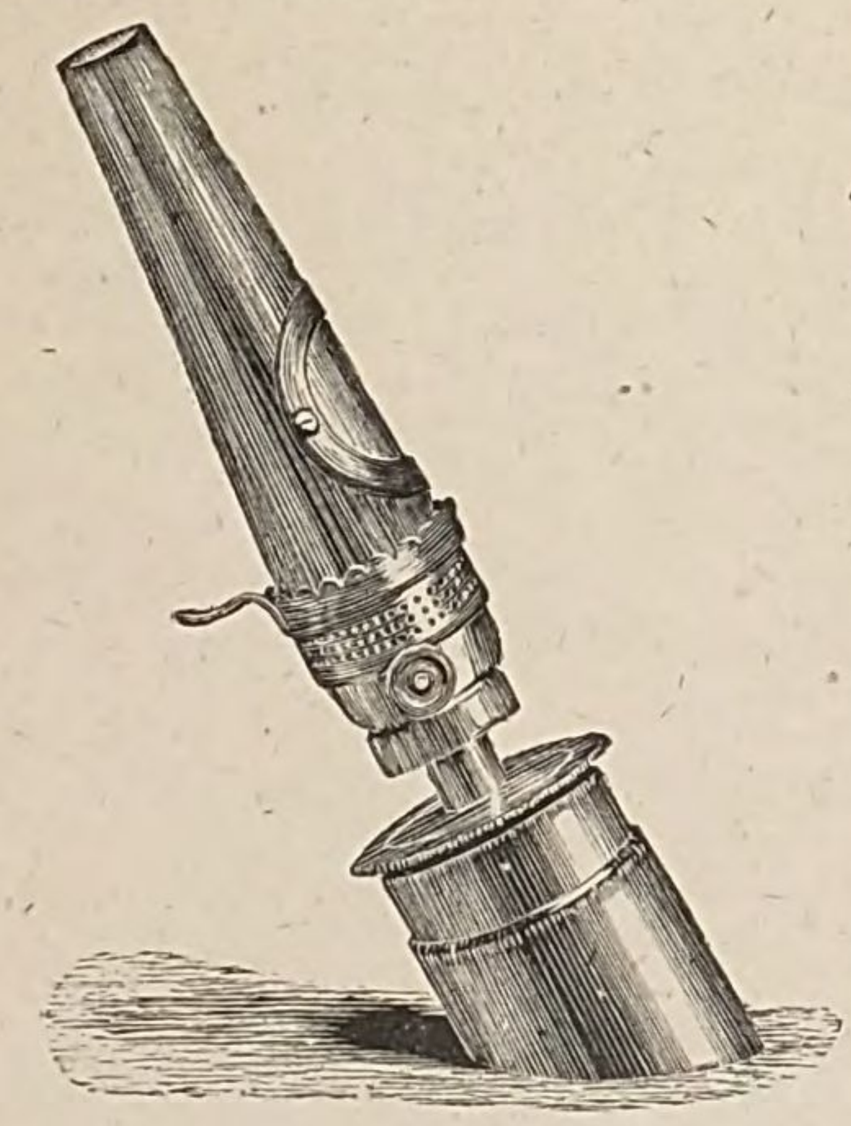
The lamp which I find upon the whole most advantageous for microscope work, even when the 1-50th of an inch object-glass is used, is one of the small paraffin lamps with a round wick, which can now be purchased for 1*s.* 6*d.* I recommend every one, even if provided with gas, to obtain one of these small paraffin lamps. Different forms are represented in figs. 1, 3, 5, pl. IV. The form I prefer is the little one seen in figs. 1 and 2, pl. IV. This is so small that it can be carried about very easily and placed very near to the object, a desideratum in some cases. The figures represent the lamp one-third of the real size. It was made for me by Mr. Swift, but other instrument makers now provide these useful little microscope lamps. I have had a sort of shoe made for it, so that the lamp can be inclined, as represented in

Fig. 1.



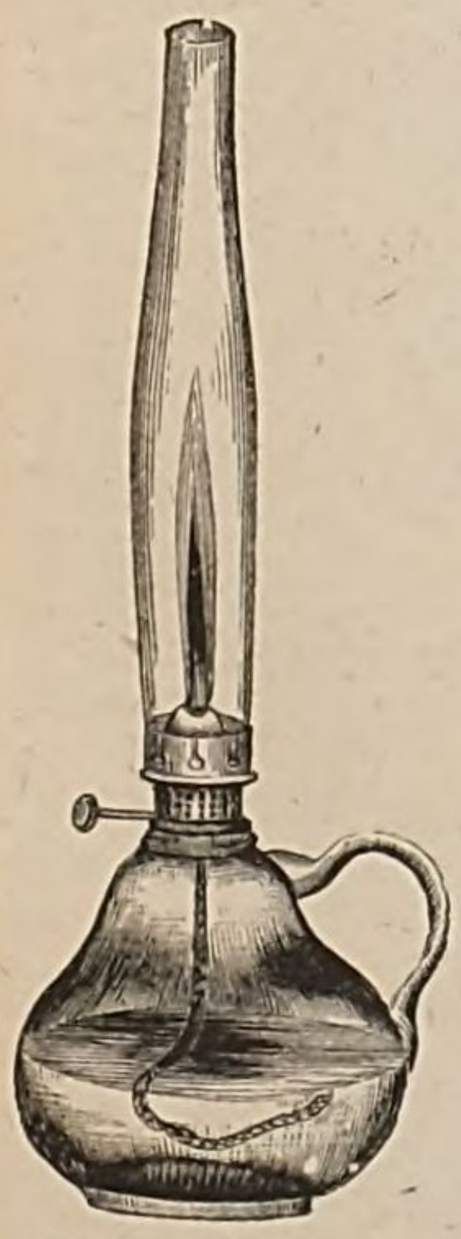
Round-wicked paraffine lamp,
½ real size. p. 26

Fig. 2.



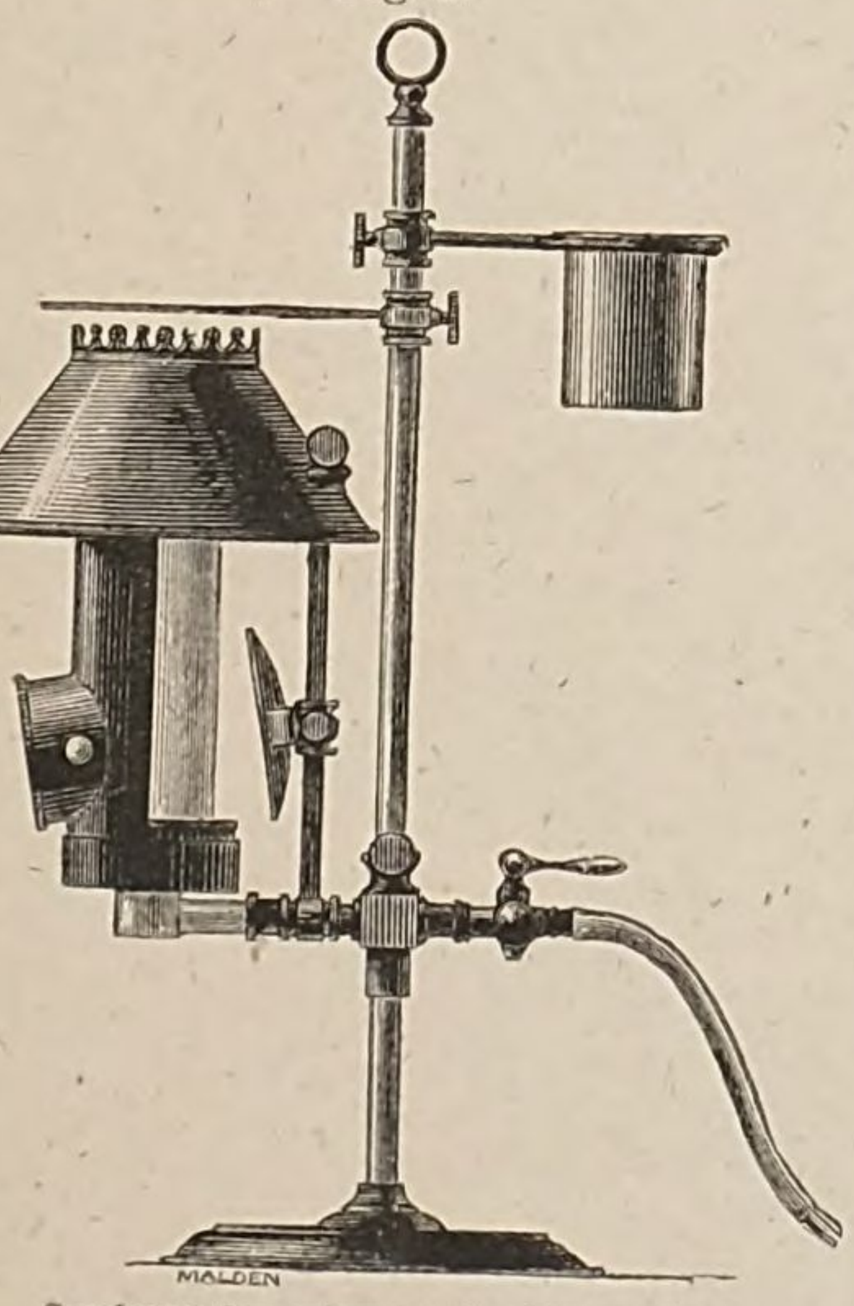
The same as fig. 1, inclined in its shoe p. 26.
See also Fig 1. Plate I.

Fig. 3.



Simple paraffine lamp,
with round wick. p. 26.

Fig. 4.



Gas lamp for microscopic observation,
arranged by Mr. Highley. p 26.

Fig. 5.

Bockett lamp, made by Collins
with round wick. p. 26.

Fig. 6.

Dr. Maddox's spring clip, for pressing thin glass
cover while the cement is drying. p 28.

Fig. 7.

Modified spring clip, for keeping thin glass cover
in its place. p. 28.

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fig. 2, and the flame made parallel to the lens of the condenser, by which the most satisfactory illumination may be obtained for the highest powers. See pl. I, fig. 1.

Apparatus for Drawing and Measuring Objects, and for ascertaining the Magnifying Power of Object Glasses.

9. Neutral Tint Glass Reflector.—This fits on the eye-piece, the microscope being arranged horizontally.—H. to W., § 44.

10. Common Hard Pencils, *steel pens, Indian ink, fine Bristol board, tracing paper, retracing paper.* To be obtained of Mr. Brodie, Artist's Colourman, Long Acre, near Drury Lane.

11. Stage Micrometers *divided into* 100*ths and* 1000*ths of an inch.* —H. to W., § 60.

Instruments and Apparatus for General Purposes.

12. Wire Retort Stand for supporting watch-glasses, &c., pl. VI, fig. 1.—H. to W., § 70.

13. Tripod Wire Stands, pl. VI, fig. 2.

14. Spirit Lamp, pl. VI, fig. 1.—H. to W., § 69.

15. Evaporating Basins, pl. VI, fig. 2.

16. Watch-glasses.

17. Thin glass, cut in squares and circles.

18. Plate Glass and Common Glass Slides, all three inches by one inch. No other sizes should be used.

For making Dissections, and for Cutting Thin Sections of Soft and Hard Tissues.

19. Common Scalpels. Double-edged Scalpel, pl. VII, fig. 1.

20. Valentin's Knife, pl. VII, figs. 2, 3.

21. Scissors, ordinary form, and a small pair with curved blades, pl. VI, figs. 4, 5, 6.

22. Needles, mounted in handles, for dissecting, pl. VI, fig. 3. The handles of crochet needles are convenient for holding the needles, some of which may be flattened near the points, so as to serve for delicate knives.

23. Forceps.—One pair of ordinary dissecting forceps, and one pair with curved blades, pl. VI, fig. 7. **Forceps for sputum,** fig. 8.

24. Glass Dishes, of various sizes, from an inch to two inches in depth, for dissecting under water.

25. Loaded Corks, pl. VII, figs. 4, 6.

26. Fine Pins.

27. Saw, with fine teeth, for cutting thin sections of bone and other hard tissues, pl. VII, fig. 5.

28. Hones, for grinding sections of bone thinner, and for polishing them.

29. Strong Knife or Razor for cutting thin sections of bone, &c. The razors that can be purchased for a shilling each answer well. They may be fixed in a strong handle.

Cements, Preservative Fluids, and Apparatus for Mounting Objects in Air, Aqueous Fluids, and Balsam.

30. Brunswick Black, containing a few drops of a solution of India-rubber in coal naphtha. **Gold Size**, or **Bell's Cement** for glycerine preparations. H. to W., § 90, 91. **Gum Damar** dissolved in Benzole answers well.

31. Spirit and Water.—H. to W., § 99.

32. Glycerine (Price's).—H. to W., § 100.

32.* Potassium Acetate (Saturated Solution) for tissues which would be rendered too transparent by glycerine.

33. Gelatine and Glycerine.—H. to W., § 106.

34. Solution of Naphtha and Creosote.—H. to W., § 102.

35. Carbolic Acid and Water.

36. Chromic Acid.—H. to W., § 104.

37. Turpentine. Canada Balsam. Gum Damar. Oil of Cloves. Canada balsam is soluble in ether, chloroform, turpentine, and benzole.

38. Cells of various sizes.

39. Small Glass Shades, *to protect recently mounted preparations from dust*, pl. VIII, fig. 1.

39.* "Spring mounting Clips," pl. IV, fig. 6 & 7.

For the separation of Deposits from Fluids.

40. Conical Glasses, pl. XV.

41. Pipettes, pl. XVI, fig. 5.

42. Wash-bottle, pl. XVI, fig. 6.

43. Animalcule Cage, pl. XVI, fig. 10. **Glass Cell**, fig. 9.

For making Injections.

44. Injecting Syringe, holding from half an ounce to an ounce, pl. VIII, fig. 4, or the apparatus represented in fig. 3.

45. Pipes of various sizes, pl. VIII, fig. 5.

46. Corks for stopping the pipes, pl. VIII, fig. 6.

47. Needle for passing the thread round the vessel, pl. VIII, fig. 9.

48. Bull's-nose Forceps, for stopping vessels which have been divided, pl. VIII, fig. 8.

49. For making Blue Injection.—*Ferrocyanide of potassium.* "*Muriated tincture of iron*," *or the* "*Tincture of perchloride of iron*,"

of the British Pharmacopœia. Ferridcyanide of potassium and sulphate of iron, for making *Turnbull's blue*, a modification of the Prussian blue. *Glycerine and spirits of wine* for preparing the *blue injecting fluid* (§ 97).

49*. For making Carmine Injecting Fluid.—Best carmine. Strong liquour ammonia. Parchment size.

Chemical Analysis in Microscopical Investigation.

50. Platinum Foil. Platinum Capsule. Wire.

51. Test Tubes and Rack.

52. Small Tubes, about an inch or an inch and a half in length, fitted with good corks, to preserve pieces of tissue, deposits, &c., in glycerine and other media.

53. Stirring Rods.

54. Evaporating Basins. Watch-glasses.

55. Small Glass Bottles with capillary orifices.

56. Wire Triangles, tripods. **Small Retort Stand.**

57. Small Flasks.

58. In small Half-drachm Bottles:—Crystals of nitrate of silver. Strong solution of chloride of gold. Osmic acid. Pure common salt. Chloride of calcium. Chromic acid. Bichromate of potash.

Reagents in One or Two Ounce Stoppered Bottles.

59. Distilled water.

60. Ether.

61. Nitric Acid.

62. Acetic Acid.

63. Ammonia.

64. Solution of Potash.

65. Solution of Soda.

66. Nitrate of Silver.

67. Nitrate of Barytes.

68. Oxalate of Ammonia.

69. Iodine Solutions.

70. Chloride of Calcium.

71. Acetate of Potash.

72. Test Papers.

The student will find the little drop bottles described in Chapter X, and figured in pl. XVI, very convenient, and as they are inexpensive, he might be provided with a few for the tests in common use.

*** The instruments and apparatus above enumerated, may be obtained of Mr. Hawksley, Blenheim Street, Bond Street; Mr. Baker, Holborn; Mr. Swift, 43, University Street, Tottenham Court Road, who will take trouble to procure anything the student requires. They are also furnished by most instrument makers. Many have been figured in "How to Work with the Microscope," and figures of some of the most important have been repeated in the plates in this work.

General Remarks on Examining Objects and on Recording Microscopical Characters.

It is not within the limits of the present work to describe the anatomy of tissues in a healthy and morbid state, but I shall refer to those methods for demonstrating the anatomy of healthy and diseased structures, which, according to my experience, are the most useful. In cases in which any particular method of investigation is required, I shall, if possible, give illustrations of its use. The student will, I hope, by occasional reference to other works, be enabled without much difficulty to fill up for himself the deficiencies of the present volume.*

73. Method of submitting a portion of Tissue, or other Object, to Microscopical Examination.—Transparent objects may be examined by *transmitted* and by *reflected light*. By the former we learn the nature of the texture and internal arrangement of tissues, while by the latter mode of examination we can only recognize peculiarities of the surface.

For examination by transmitted light, an object must be sufficiently thin and transparent to permit light to pass through it readily, pl. I, fig. 1, while thickness and opacity present no impediments to examining its surface by throwing the light down upon it (reflected light). —See H. to W., § 23 to § 29. Objects may be examined with very high powers by reflected light with the aid of a very ingenious arrangement, which can be adapted to any good microscope.—See H. to W.

In order to subject a portion of tissue or any substance to examination by transmitted light, one usually proceeds as follows :—A glass slide is carefully cleaned, and the thin section of tissue which has been removed by the aid of forceps and scissors, or a scalpel, placed in the centre ; a drop of clean water, serum, weak glycerine, or other liquid, is then added, and the whole covered with a clean square or circle of thin glass. The thin glass is to be allowed to fall gently on the specimen, one edge being first brought in contact with the fluid, and the surface being gradually wetted as it is allowed to cover the specimen. If the under surface of the thin glass be gently breathed upon, it becomes wetted more easily. The specimen may be teased out with needles, pressed, or unravelled, if necessary, before being covered with the thin glass. If the substance be covered with much soft pulpy matter, or débris produced in the process of cutting the section, it may be slightly washed in water before being placed upon the slide, or a jet of water from the wash-bottle may be forced upon it. The fluid in which the specimen is immersed is kept from dust, and very conveniently applied by the

*"Quain and Sharpey's Anatomy." "Kölliker's Anatomy." "Stricker's Anatomy." "Frey on the Microscope." "The Archives of Medicine," and Papers by the author in "The Phil. Trans." "The Physiological Anatomy and Physiology of Man." "The Structure of the Tissues." "Bioplasm : an Introduction to Physiology and Medicine."

aid of the little bottle figured in pl. VIII, fig. 2. Thin sections will require to be laid flat upon the slide, with the assistance of needles and forceps, or may be manipulated with fine camel's hair pencils.

74. Of the Media in which Objects should be Examined.—With reference to the medium in which any particular object is to be examined, but few rules can be laid down. Many structures may be examined in water, but it should be borne in mind that this fluid often alters the character of the tissue very much more than many other fluids which it has been theoretically assumed are unsuitable.

Generally, tissues should be submitted to examination in a medium which closely resembles that which surrounds them during life in density and fluidity. Thus, albumen and water form a very useful fluid for examining many structures. In a fluid of this kind, made to resemble as closely as possible in density and in chemical composition the medium which bathes the tissues during life, we may conclude that the appearances observed are natural, and not produced artificially. There are, however, many cases in which it is desirable to examine a tissue in a medium of much greater density than that with which it is ordinarily surrounded. Highly refracting structures may require immersion in a highly refracting material before their arrangement can be made out. When a section of a tissue appears thick and opaque in water, immersion in such a medium often renders it perfectly clear and transparent. White fibrous tissue, although so opaque, even in very thin layers, as to prevent structures embedded in it from being seen, may be made perfectly clear and transparent by being immersed in ordinary syrup, clear and almost colourless treacle, strong solution of grape sugar, or in glycerine. Of these fluids, glycerine is the most convenient, and can be easily diluted to any required strength. When employed dilute, it is well to place a piece of camphor in the bottle in which it is kept, which prevents it from becoming mouldy. In the investigation of morbid growths, great advantage will be gained by the use of glycerine, but when fibrous tissue is present, its characters must be studied in water, or in some aqueous fluid of very moderate density. When observing the appearances of a structure in glycerine, allowance should always be made for the greater transparency of the fibrous tissue. Ever since I began work I have been well acquainted with the value of glycerine, and have never failed to advocate its use. After years of opposition, based upon fanciful objections, it is satisfactory to find that the advantages of this medium are now becoming generally recognized. The composition and methods of using different preservative solutions are fully discussed in "How to Work with the Microscope," § 19 to § 113. The methods of preparing and mounting objects in glycerine and other media will be found described in Chapter III, p. 51.

75. Of making and recording Observations, and of drawing In-

ferences from Microscopical Appearances.—The difficulty of making out the structure of many organs and tissues is great, and very considerable practical experience will be required before the anatomical characters of a healthy texture can be distinctly demonstrated by the student. These difficulties are much increased in the examination of morbid growths. When chemical reagents are applied to ordinary tissues, the effects must be very carefully observed, otherwise there is danger of mistaking the change of character produced by the application of the reagent, for a morbid alteration. Even the addition of a drop of water often materially alters the microscopical characters of a tissue.

It is only by very frequent and careful examination of morbid growths, that the observer can hope to recognise and interpret their characteristic appearances, and it should only be with the utmost caution, and after long familiarity with microscopical examination generally, that he should attempt to pronounce an opinion with reference to the nature of a morbid growth. Without extensive observation and great care, he will run the risk of bringing discredit not only upon himself as an observer, but also upon microscopical investigation generally.

The opinion is much too common that a good instrument and the necessary apparatus are alone required to make a microscopical observer, and it is well that every one should guard himself at the outset against so serious a mistake. Each one must educate his eye for himself, and although he will undoubtedly receive some assistance from the teaching of others, from books and faithful drawings, he must not depend upon these, but must rely chiefly upon his own experience and perseverance. No one who does not at once make up his mind to give up much time to the pursuit, can ever become an accurate observer, educate himself so as to form a correct judgment, or learn how to avoid making great mistakes ; and he who cannot, or is unwilling to spend considerable time in work, had better not take up microscopical enquiries. A good knowledge of drawing, of the stethoscope, of the opthalmoscope, and indeed of any other mode of investigation accessory to medical research, requires far more devotion than is implied in the mere sacrifice of the money which is necessary for the purchase of books and instruments. So it is with the microscope ; and he who has the largest means at his disposal for obtaining the most costly instruments made, and all the books published, and with the advantages of the best tuition, is less likely to become a useful, earnest labourer in this field of enquiry, than the student who spends his four or five pounds in a simple instrument, without any luxurious accessories—but having a conviction that the study is real, and worthy of attention, and a determination to set to work honestly and zealously with the hope of being one day able

to add his work to that of men who have worked before him, whose lives and labours he respects and honours.

The student is recommended to examine very frequently the structure of the kidney and liver in man and many of the lower animals in health, because these organs are very often the subjects of investigation in cases of disease; the changes in structure which they undergo having received a large share of attention.

Of Drawing, Engraving, and Measuring.

76. Of Drawing Objects.—It may almost be said that all progress in our knowledge of minute structure, both as regards healthy and diseased tissues, depends upon the drawings which are made. It is almost hopeless for an observer to attempt to describe what he sees in words; and such descriptions, however detailed they may be, cannot, with any advantage, be compared with those of others. It is often asserted that an observation recently made by B, has been anticipated by A, when, upon careful reference, it turns out that A never had the remotest conception of what was discovered by B. But a truthful drawing of what a man has seen recently, may be compared with drawings which may be made a hundred years hence, although the means of observation will be far more perfect than any that we can at present employ. Much will be learned by such comparisons. I am sure that an honest enquirer cannot be of greater use in his time than by making good drawings of what he has seen;—these will be of far greater help to our successors, than any amount of description we can write for them, and we may feel sure that they will look at our drawings if they are honest copies of nature, while we all know that comparatively very little of what we write will be read when the whole aspect of this and of every other department of science shall be changed.

Whatever is observed is worth copying, provided it has not been correctly copied before. Much remains to be done in representing microscopic texture faithfully. Photography has been of advantage, and will doubtless assist more; but there are many structures, the colour of which alone renders it quite impossible to obtain photographs of them.

It can only be by patient study that any one can hope to be able to render accurately by hand the beautiful and delicate lines and tints in many microscopic objects; but it is so important that this should be done well, that I cannot too strongly urge all those who wish to work at the microscope, earnestly to practise drawing as much as possible. On micro-photography, see H. to W., Chapter IX, p. 149 to p. 189.

It is beyond the power of language to describe the characters of many structures in such a way that their appearance could be repro-

duced in the mind of another; and even if this could be done, so wonderfully delicate and minute are the observed differences in many cases, that any attempt to classify and arrange our observations would seem to be at present hopeless, and must become more hopeless in proportion as observations multiply. The different meanings which different persons attach to words and phrases, give rise to another difficulty in an attempt to collate and deduce inferences from the observations which have been made. It therefore seems to me that all advance in our knowledge of structure, as well as of the minute changes incessantly going on in living organisms, really depends upon accurate copies of the objects being made. In this way alone can the work of the present generation be useful to that which succeeds it.

In delineating an object magnified by the microscope, it is important to copy it correctly, both as regards the relative position of the several structures to each other, and also with respect to their dimensions. To copy the size exactly will be found extremely difficult by the eye alone, but there are several ways of proceeding by which accuracy may be ensured. Some of these I shall now briefly describe. The simplest method is to place the paper upon the same level as the stage upon which the object is situated. If we now look steadily at the object with one eye, while the other is employed to govern the movements of the pencil, the object will appear to be thrown as it were upon the paper, and its outline may be very readily traced. By a little practice, the relative size of objects may be ensured in this manner, but it is troublesome and difficult to keep both the object and paper perfectly steady.

The principle of the camera lucida has been applied to taking microscopical drawings, and has been found to succeed admirably. The object appears to be thrown down upon the paper, and with a little practice the observer may trace the lines with great accuracy. If a little *steel disc* smaller than the pupil of the eye be placed at an angle of 45° with the eye-glass, it will receive the magnified image of the object and reflect it upwards upon the retina of the observer. The simplest and cheapest reflector for microscopical drawing, consists of a small piece of plate glass slightly coloured, in order to improve its reflecting power, but still not so dark as to prevent an object being seen through it perfectly.—H. to W., § 44, pl. XVII. Mr. Kesteven uses an ordinary thin glass square or circle.

77. Of Drawing with these Instruments.—In order to use the above instruments, the microscope is arranged horizontally, and the paper placed on the table. It is important to arrange the light very carefully. The image should not be illuminated too intensely, and the paper upon which the drawing is made should not be too much in the shade, or the point of the pencil will not be seen distinctly. Experiment can alone decide the relative intensity of the light upon the object and upon the

paper, but with a little practice the proper amount of illumination will be discovered. The distance between the reflector and the paper should be precisely the same as from the object to the eye-piece, for otherwise the size of the object delineated will be altered, or an arbitrary distance of ten inches may be always adopted.

The object appears to be thrown upon the paper, and its outline may be very readily traced. If it is to be drawn smaller, it is only necessary to place the paper upon a stand closer to the reflector. If, on the other hand, a large *diagram* is required, the distance must be increased. By placing the diagram paper upon the floor, the object can be readily traced with a long pencil. In this manner many of my diagrams have been made. They must of course be accurate copies of the objects themselves, and are therefore far more truthful than diagrams copied from drawings representing microscopical structure, can be. If the distance of the diagram paper be always the same, the drawings so obtained may be compared with each other, and scales of measurement obtained by magnifying lines of the stage micrometer in the same degree may be appended to them.

78. Of making Drawings which it is intended should be Engraved. —With a little practice, the observer may acquire the art of drawing on wood, and the engraver will often be able to produce a more faithful representation of the object than he could do if he copied from paper the *drawings* made by the microscopical observer. It is, however, necessary to practise the plan of producing varieties of tints, by straight lines, whenever this can be done, as the labour of engraving is thus much economised. The drawing should first be made roughly on paper, in order to obtain the size and general characters of the object. A piece of retransfer paper is then placed between the prepared block and the paper sketch, and the prominent lines of the drawing traced with some blunt-pointed instrument (a needle, the point of which has been made blunt by filing it, answers very well). By using a slight pressure, the colour of the retransfer paper is transferred to the wood block in the lines corresponding to those of the drawing. These lines are afterwards traced with lead pencil, corrected, if necessary, and the delicate parts of the drawing filled in by carefully copying from the object.

If the engraving is to be a facsimile of the drawing with the different parts in corresponding places, it is necessary, in the first place, to copy the picture with ordinary tracing paper, and *invert* the tracing upon the retransfer paper on the wood block, as the impressions are of course always reversed; or a reverse may be obtained by copying the image of the drawing reflected from a looking-glass.

79. Tracing Paper, Retransfer Paper, and Wood Blocks can be obtained all ready for use at the artists' colourmen. See list of addresses at end of the volume.

80. On Measuring the Diameter of Objects.—In giving descriptions of microscopical characters, it is well to refer the reader to properly arranged scales appended to every drawing, instead of alluding to the dimensions of an object in the text. If these scales are magnified in the same degree as the objects delineated, the diameter of every object depicted, may be at once read off. This plan has been followed in all my papers and books. At the foot of each page of drawings a scale is appended. For all ordinary purposes, it is only necessary to compare roughly the size of the drawing with the scale, which is magnified in the same degree as the specimen itself, but in those instances where great accuracy is important, a pair of compasses may be used. In my memoirs, I have not stated the dimensions of any of the objects, because any one can readily ascertain these for himself, by reference to the scales appended.

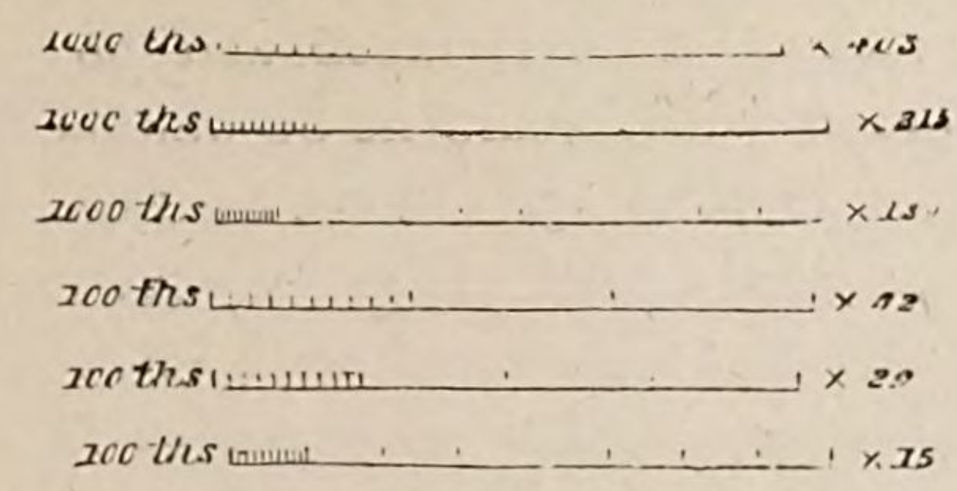
I cannot therefore too strongly recommend all microscopic observers to ascertain for themselves *the magnifying power of every object-glass*, and to prepare, as described in the two following sections, *a scale of measurement by which the dimensions of every object can be at once ascertained.*

81. Mode of ascertaining the Magnifying Power of the Object-glass.—A glass micrometer divided into 100ths of an inch is placed in the focus of the object-glass of the microscope, which is arranged horizontally. The neutral tint glass-reflector is fitted to the extremity of the eye-piece, and the light carefully adjusted so as to render the micrometer lines distinctly visible. Care must, however, be taken that the distance from the object-glass to the reflector is the same as from the latter to the paper beneath it, upon which the magnified micrometer lines may now be traced, unless the arbitrary distance of ten inches from the eye-piece is invariably adopted. A four or six-inch scale, accurately divided into 10ths of an inch, is now applied to the magnified 100ths of an inch, which have been traced on paper, and the magnifying power of the glass is at once ascertained. Suppose each magnified 100th of an inch covers one inch, the magnifying power will be 100 diameters: if an inch and three-tenths, 130 diameters; if four-tenths of an inch, 40 diameters; and so on, each tenth of an inch corresponding to a magnifying power of ten times.

If we wish to ascertain the magnifying power of one of the higher object-glasses, a micrometer divided into 1000ths of an inch should be employed instead of the one just alluded to. In this last case, each tenth of an inch upon the scale, corresponds to a magnifying power of one hundred instead of ten diameters. Any fractional parts can be readily estimated if we have a very accurately divided scale. This process must be repeated for every object-glass, as well as for each different eye-piece employed with the several objectives.

82. To ascertain the Diameter of an Object.—If an object be substituted for the micrometer, and its outline carefully traced upon paper, its dimensions may of course be easily ascertained by comparison with the micrometer lines. The magnifying power used being the same in both cases.—H. to W., § 64.

The following are examples of some of the scales referred to :—



1000ths and 100ths of an English inch magnified in various degrees.
The smallest divisions indicate 10,000ths and 1000ths of an inch.

83. Standards of Measurement.—In this country we usually employ fractions of an English inch, but on the continent the Paris line = ·0888, or about 1-11th of an English inch, is very generally used. The sign ‴ is used to signify "of a line," and has been employed by Professor Kölliker in his works, while ″ signifies "of an inch." In order to compare the researches of different authors, it is often necessary to convert one expression of measurement into another.—See H. to W., § 66.

84. Of Finders.—Several different plans have been employed for the purpose of finding any special object upon the glass slide, or any particular part of a specimen without loss of time.—See H. to W., § 67, pls. XVIII, XIX. But a very simple and efficient method has been proposed by Mr. Bridgman, of Norwich, and made by Mr. Baker, of Holborn. Attached to the side of the body of the microscope is a brass arm capable of being moved upwards and downwards in such a manner that it may fall upon one end of the slide. If the pointed end be anointed with ink or black varnish, a mark may be made upon the end of the slide when the point for observation is in the centre of the field. In order to find this same spot at any future time, it is of course only necessary to place the slide in such a position that the original mark exactly corresponds with the point of the finder, and the part of the specimen must then be again in the centre of the field. The plan is so simple and efficacious, that it will, I think, completely supersede the various finders now in use.

Professor Rutherford adopts the following plan in commencing his class demonstration. (See "Notes of a Course of Practical Histology for Medical Students.") "A simple object like yeast is taken first. I give no description of torula, but I ask the student to describe and then to draw what he sees. Any one may be called upon to do

this. Any one who disagrees with any statement is asked to do so, and to give a demonstration in support of his opinion. To facilitate such descriptions every student has a card on which are printed the following points:—1. Shape. 2. Edge. 3. Colour. 4. Transparency. 5. Contents. 6. Size. 7. Effects of reagents. The card prevents the student from getting bewildered, and teaches him method and thoroughness. Care is taken that no one ever becomes idle. If his preparation is made, and he be waiting for his neighbours, he occupies his time in drawing. When we come to complex structures, such as bone, I give a brief preliminary account of the subject, in order that every one may understand what he sees. By questioning the student as the demonstration goes on, it is easily ascertained whether he understands what he is about. On all occasions, however, I endeavour to make the student describe what he sees. This method really *educates* him in a way such as no other method, in my opinion, can. When necessary I show preparations which have been previously made. With a subject like teeth this is, of course, necessary, but whenever it is possible each student makes his own preparations, and preserves them when they are worth keeping. Every student provides himself with a box of slides and cover glasses, scissors, forceps, scalpels, razor, mounting clips, needles in handles, camel-hair pencils, while he is furnished with all the tissues, reagents, &c. I give every student a stand with twelve one-ounce bottles, containing the reagents. The bottles have good corks, in which are fixed glass rods, sable, or camel-hair pencils. Some may say that corks are bad in such a case. If expense were no object, of course one would use Beale's excellent drop bottles, but they are too expensive (?) for class purposes, and really the corked bottles do very well. I very rarely find a single cork cell in any preparation. [The bottles in question may be obtained for 6*d.* each. See Chapter X. The bottles figured in pl. XVI, figs. 2, 3, are also exceedingly useful. They cost 1*s.* each.]

"The fluids contained in the bottles are—

"1. Distilled water, so that a drop of clean water may be had when necessary. 2. Solution of chloride of sodium 0·75 per cent. (7·5 grains dissolved in 1000 grains of water), for treating delicate protoplasmic tissues. This is commonly called salt solution. 3. Absolute alcohol. 4. Oil of cloves. 5. Oil of turpentine. 6. Glycerine. 7. Acetic acid. 8. Weak spirit (1 part methylated spirit, 3 parts distilled water). 9. Solution of magenta for staining. 10. Saturated solution of potassium acetate for preserving osmic acid preparations. 11. Solution of Canada balsam in turpentine. (Pure Canada balsam, dried till it is hard and crystalline, and then dissolved in turpentine.) 12. A thick solution of Damar resin in benzole, as a luting."

CHAPTER II.

Methods of Examining Tissues.—Preliminary Operations.—Of Hardening Tissues.—Of Freezing Tissues.—Of Washing and Pressing and Drying Tissues.—Igniting in order to remove Organic Matter.—Of Softening Tissues by Artificial Digestion.—Of rendering Tissues more Opaque or more Transparent by Chemical Reagents such as Alkalies and Acids.—Lockhart Clarke's Method of Preparing the Brain and Spinal Cord.

In order to examine the structure of many tissues, it is necessary to obtain a section evenly cut, and sufficiently thin to allow plenty of light to be readily transmitted. The difficulty of making thin sections of many textures is often very great, and the section, if required more than a quarter of an inch in diameter, cannot be readily made with an ordinary scalpel. Sometimes we require to cut a thin section of a texture so soft that it can scarcely be touched without injuring its delicate structure and the position of its constituents being altered; while, in other instances, we have to obtain a very thin transparent section of a substance so hard, that steel tools will scarcely scratch it, as the enamel of teeth, fossil teeth, &c.

Before the operation of cutting a thin section can be performed, it is sometimes necessary to soften the tissue by soaking it in some chemical solution. In other cases, the texture requires hardening, in consequence of being too pulpy and soft to be cut with a knife. A tissue may be hardened in some cases by drying it, but various chemical substances are highly valuable for this purpose.

Preliminary Operations in Examining Tissues.

85. Of Hardening Tissues by Boiling.—This operation is often of great service in enabling us to demonstrate the structure of a tissue which in its natural state is too soft or pulpy to manipulate. For instance, the fibres of which the crystalline lens is composed, are best shown after boiling the lens in water. They may be separated from one another by being boiled in a 1 per cent. solution of sulphuric acid. The branched muscular fibres in the tongue of the frog, in the heart, and in other situations, may be made out very readily by boiling the tissue in water for a few moments, and then tearing up small portions

with fine needles. Beautiful sections of muscular fibre can often be obtained after the texture has been boiled in water. Various glands and other tissues often require to be boiled for some time in water, in order to harden them. In all cases the microscopical characters of the recent texture should be ascertained, as well as those of the tissue which has been hardened by boiling. Small portions of tissue can be readily boiled in a test-tube over the spirit-lamp.

Of late years this operation has for the most part been replaced by other methods of hardening.

86. Of Freezing Tissues.—Permanent hardening by the application of heat has given way to temporary hardening by congelation. If soft textures be frozen with care, they may be made sufficiently firm to cut excessively thin sections which may then be allowed to thaw and examined in the usual manner. We may operate upon perfectly fresh tissues in this way. More than 25 years ago I remember Mr. Bowman made sections through all the tissues of the eye after the organ had been frozen, and in Russia the plan of freezing has been long in use both for the purpose of ordinary dissection and for the more delicate operations required by the microscopical observer. The freezing mixture usually employed is pounded ice and salt, but many other methods may now be adopted. A piece of tissue may be instantly frozen by placing it in gelatine, or in white of egg and water in a small vessel which can be placed in a test tube, upon which a jet of nitrous oxide gas is allowed to play for a few seconds. A tube with a fine opening must be properly fixed in one of the iron bottles in which the gas is condensed. These are now prepared in great numbers for the dentists for the purpose of inducing anæsthesia. Carbonic acid answers equally well, but as the nitrous oxide is now prepared in large quantities, there is no difficulty in obtaining one of the iron bottles at any time. This means of freezing may also be adopted in using the section cutter described in § 112, but the greatest care must be taken not to allow the jet of gas to come near the fingers, for the process of congelation acts very quickly and severe frost-bite would be the result. Nor must the metal pipe through which the gas is issuing be touched.

87. Drying the Tissue previous to Examination.—Thin sections of certain tissues may be obtained by drying the substance thoroughly in the first place; and then cutting off a thin shaving with a sharp knife. In this way, specimens of skin, mucous membrane, and many other tissues, are advantageously prepared. The tissue is stretched on a board with pins, and then allowed to dry, when a very thin section can be cut off and examined in Canada balsam; or it may be soaked in water for a short time, and immersed in glycerine or other fluid. When subjected to examination, it will have regained its fresh appearance. Portions of muscular fibre, pieces of the tongue, skin, the sclerotic,

choroid, and retina, and many other tissues, may be allowed to dry in this manner, and with a sharp knife exceedingly thin sections obtained, which could not be procured in any other way. The drying may be effected in a warm room, or in a current of air. A high degree of artificial heat should be avoided, and in many cases the best plan of drying a tissue is to place it in a basin under a bell jar, supported on a piece of coarse wire gauze, over sulphuric acid. The process is expedited by exhausting the air, which may be readily effected under the receiver of a small hand air pump.

88. Of Washing, Soaking, or Pressing Tissues.—Not unfrequently it is necessary to get rid of the soft and more pulpy part of a tissue, in order to subject the more dense and fibrous portion to examination. This object is usually effected by soaking the tissue in water for some little time, and then placing it under a running stream of water, by which means the softer portions are gradually washed away. Soaking in water frequently enables us to tear up a tissue very readily with the aid of needles, and thus to demonstrate its structure. Occasionally it is found necessary to press the tissue, and rub parts of it together, before the soft pulpy portions can be got rid of. In this way we may demonstrate the supporting or trabecular tissue of the spleen, and the areolar and vascular tissue of many organs. Thin sections of kidney, liver, and other glandular organs, may be thus treated when we wish to wash away the epithelium and blood, in order to study the characters of the tissues which remain. In these operations the wash-bottle, pl. XVI, fig. 6, will be found useful. Generally it will be better to make a thin section of the tissue first, and then soak and wash carefully, when the parts may be seen *in situ*. The process of rendering delicate tissues extremely thin by pressure is greatly facilitated if the tissue be immersed in a viscid medium such as strong glycerine or syrup instead of water. In this way the tissue may be frayed out without being destroyed or crushed, and thus its structure may be most clearly demonstrated.

89. Igniting the Substance in order to remove Organic Matter.—When the inorganic part of a tissue which is not altered by exposure to a red-heat is to be examined, recourse may be had to ignition, in order to get rid of the animal matter. In this way, crystals of carbonate and phosphate of lime, and granules of siliceous matter may be separated from the organic material with which they were combined. The beautiful siliceous shells of the diatomaceæ may be thus obtained. The ignition should be performed in a small platinum capsule, supported on a tripod (pl. VI, fig. 2), or upon a small piece of platinum foil. The carbonaceous residue must be exposed to the dull red-heat of a spirit-lamp for some time, until only a pure white ash remains, which will be found to contain the objects of our search in a very perfect state. If the siliceous matter only is wanted, the ash should be treated

with strong nitric acid, which will dissolve any carbonate or phosphate. The insoluble residue may then be washed and dried, and subjected to microscopical examination, immersed in water, glycerine, turpentine, or Canada balsam. In many cases, this method is superior to that of boiling in nitric acid, in order to remove the organic matter. Both processes may, however, be employed where only the siliceous residue is wanted; but if we require the calcareous salts, ignition at a dull red-heat is alone applicable.

90. Of Softening Tissues by Artificial Digestion.—This plan is of great use in the preparation of many very delicate tissues which are to be examined under the highest powers. I used it a good deal in 1864, when studying the arrangement of the nerves in the papillæ of the frog's tongue. By the digestive fluid the fibrous tissue is softened, and nerve fibres and other delicate textures can be more readily traced. Moreover, the consistence of the half-digested section is such that it may be most advantageously subjected to pressure.

The fluid is made as follows:—Five grains of pig's pepsine are well mixed with an ounce of distilled water, and two drops of strong hydrochloric acid added. After the mixture has been kept at the temperature of 100° or thereabouts for an hour, it is carefully filtered.* One part of the clear digestive fluid is mixed with two parts of strong Price's glycerine. The sections of tissue to be softened are placed with a few drops of the glycerine digestive fluid in a small tube and kept at the temperature of 100° for one, two, or three hours. They are then to be transferred to plain glycerine or glycerine rendered acid by acetic acid (page 66) and examined in the usual manner. The digestive fluid and glycerine will keep for any length of time. It may be made much stronger if desired. The student who adopts this method of investigation will soon discover many useful modifications of the plan I have recommended.

91. Of rendering Transparent Tissues more Opaque, and of making Opaque Tissues more Transparent by Chemical Reagents.—Many tissues which are perfectly transparent, and apparently structureless when subjected to ordinary examination, can be shown to possess a peculiar structure if treated with some chemical reagent which has the property of rendering them more or less opaque. In many cases the granular appearance produced by certain reagents, depends upon the precipitation of albuminous matter. Thus a weak solution of alcohol often enables us to cause coagulation of the surface of a mass of living matter or bioplasm, and many have been led to infer from the

* This is made by Messrs. Bullock and Reynolds, 3, Hanover Street, Hanover Square. The method of preparing it is described in the "Archives of Medicine," vol. I, and also in "Kidney Diseases, Urinary Deposits, and Calculous Disorders," 3rd edition, page 277.

appearances produced, the presence of an external membrane or cell wall where no such structure really existed. Sometimes coagulation is effected in certain lines only, and in some instances, important deductions may be arrived at, concerning the structure of the texture, as well as the manner in which the new matter was deposited. Chromic acid renders some perfectly transparent structures composed of albuminous matter more or less granular, and by the action of this substance, peculiarities of the tissue which were before invisible, are often developed. The transparent vitreous humor of the eye, was shown by Mr. Bowman to possess a curiously lamellated arrangement, by the action of acetate of lead. Acids and many salts, such as alum, acetate of lead, acetate of alumina, solution of perchloride of iron, nitrate of silver, &c., effect a very important alteration in many perfectly transparent tissues.

Contrary to general opinion, many of the softest textures may however be investigated with the greatest facility after having been soaked in strong glycerine, and much concerning their structure may be learnt by this process. The glycerine used at first must be weak, and its strength must be very slowly and gradually increased. The reagents above referred to may be dissolved in the glycerine, and usually the prolonged action of weak solutions affords more satisfactory results than the quick action of strong ones. I have beautiful preparations of the most delicate embryonic tissues, preserved in the strongest glycerine. It is often advantageous to harden the tissues slightly by the addition of a little of the chromic acid glycerine solution (p. 46). (H. to W., § 299.) When once the tissues have been fully permeated by glycerine, they may be dissected and manipulated in a manner which before would have been impossible. The greatest prejudice has long existed to the use of glycerine, but it is quite certain that those who condemn it have not found out how to work with it.

Sometimes the mere addition of a coloured solution is sufficient to render a tissue perfectly distinct, which before was too transparent to be visible. A little Prussian blue, diluted with much water, or a solution of carmine in ammonia, used in a very dilute state, will in some instances enable the observer to demonstrate the presence of delicate membrane, which could not be seen before. The process of *staining* is very valuable for demonstrating delicate structural peculiarities in many transparent tissues. (See Chapter IV.)

Many structures are made perfectly clear by being immersed in certain solutions of high specific gravity, which exert no *chemical* alteration on the texture. Syrup or glycerine may be used for this purpose, but I much prefer the latter, as it is not so liable to be invaded by fungi, while it forms a most excellent preservative solution. The solution of glycerine or sugar first added should be dilute, and its strength gradually increased. This may be effected either by adding

small quantities of strong glycerine or sugar at intervals of a few hours, or by concentrating the original solution by evaporation at a gentle heat or in vacuo over sulphuric acid. White fibrous tissue, which even in a very thin layer appears opaque when examined in most fluids, becomes perfectly clear and transparent after being soaked for a short time in glycerine.

Acetic Acid renders some tissues transparent by virtue of its property of dissolving earthy salts, such as phosphate and carbonate of lime, and in other instances certain forms of albuminous matters, especially the granular matter which exists in the cell wall is made perfectly clear. Acetic acid also causes white fibrous tissue to swell up and become perfectly clear, while all traces of its fibrous appearance is lost. On all varieties of yellow elastic tissue, however, it exerts no action, so that by its use the fibres of yellow elastic tissue can be always demonstrated, although embedded in the white fibrous connective tissue.

Alkalies dissolve a great number of coagulated albuminous principles, and many opaque tissues are rendered perfectly transparent if acted upon by an alkali. The principal alkaline solutions used by the microscopist are *carbonate of potash*, *liquor potassæ*, and *liquor sodæ* (solutions of hydrate of potash and soda in water). These are employed of different strengths. They dissolve many opaque albuminous substances, if used very strong, and if diluted, render them clear and transparent. Sometimes it is desirable to render fibrous tissue transparent, in order to observe the character of certain earthy phosphates, or other substances embedded in it, which are known to be soluble in acetic acid. In such case an alkali must be employed. Instances of the application of acids or alkalies to the same end might be alluded to, but the particular advantages of one or other class of reagents will be brought forward in other parts of the work.

In some cases a texture may be dissolved, and thus other textures which were embedded in it rendered visible. The soft pulpy portion of an organ may often be got rid of by allowing a stream of water to play upon it for some time. The spleen pulp may thus be separated from the trabecular tissue of the spleen. Cells may be washed away from thin sections of liver or kidney, leaving the vessels, nerves, and connective tissue of these organs. Sometimes the addition of a little hydrochloric acid is advantageous in breaking up the cellular part of a tissue. The cellular tissue may be thus removed from the vascular and fibrous texture of leaves.

Schultze has recommended the use of chlorate of potash and nitric acid for destroying connective tissue, and Kühne has particularly advocated the process very strongly for the purpose of demonstrating the arrangement of the nerve fibres distributed to voluntary muscle, and he maintains that by this plan the nerve fibres may be shown to per-

forate the sarcolemma of the elementary fibres which are isolated from one another by the destruction of the intervening connective tissue. The solution may be made of various strengths. One or two small crystals of chlorate of potash may be placed in a test tube, and about half a teaspoonful of distilled water, and from ten to twenty drops of nitric acid, added.

The reaction with strong nitric acid sp. gr. 1·5 is decided, but if the solution is dilute, although the action will be slower, the results obtained, it is said, will be more satisfactory. The mixture may be gently warmed and freely shaken. The muscular fibres may be seen to separate from one another. The action of the reagents, especially if strong solutions be used, corrugates the nerve fibres and alters the muscles. Indeed, as far as I am able to judge, after having examined some specimens prepared by Kühne, and others made by myself according to his directions, the results are by no means satisfactory. The appearances which I have myself seen would not lead me to accept the conclusions arrived at, but it is possible that Kühne has succeeded in making specimens more distinct and definite than any I have actually seen. I think, however, that this process, as well as many other plans of preparation strongly recommended in Germany, is wrong in principle. I have proved experimentally that many of the very fine nerve fibres which I have succeeded in demonstrating by a totally different procedure, are completely destroyed or rendered invisible in tissues subjected to strong chemical actions; a much better method of effecting the same object is by artificial digestion, as described in § 90.

92. Of rendering Soft Tissues Hard and Transparent.—There are very many solutions which have the property of hardening soft tissues, but as their action depends principally upon the formation of insoluble albuminous compounds which are opaque and granular, but few are applicable for microscopical purposes. Various saline solutions as alum, bichloride of mercury, arsenious acid, &c., render most tissues too granular and opaque for observation. A very dilute solution of chromic acid of a pale straw colour, or a mixture of chromic acid and bichromate of potash, is useful for hardening many textures. In some cases a compound fluid, consisting of a mixture of two solutions—of which one has the property of precipitating albuminous substances in an insoluble state, while the other tends to dissolve them—is to be preferred. Such a solution hardens a tissue effectually, but at the same time renders it transparent. If desirable, the refractive power of such a fluid may be increased by the addition of glycerine, and with a little trouble, fluids suitable for the examination of almost every structure may be had. The solutions which I have used are the following: alcohol, glycerine, acetic, nitric, chromic, and hydrochloric acids, potash, and soda. Now alcohol, hydrochloric, and nitric acids render many

transparent albuminous textures, granular and opaque, and as is well known, produce precipitates in albuminous solutions. Alcohol will, however, dissolve fat granules. Acetic acid, potash, and soda, cause many albuminous tissues, which are more or less opaque or granular, to become clear and transparent, and dissolve insoluble precipitates of certain albuminous compounds. Glycerine, in consequence of its high refractive power, renders many tissues, which in their natural state are opaque, perfectly clear. Glycerine may be made the basis of all test solutions and preserving media. The various chemical tests may be added to it, and if the textures to be tested are well saturated with the same substance, most excellent results are obtained.

Chromic acid solution is usually employed for hardening the brain and spinal cord, but it is adapted for many other tissues as well; $\frac{1}{4}$ per cent. solution in distilled water is sufficiently strong for many tissues. Small pieces of the tissue should be placed in the fluid, left for a day or two, and transferred to fresh fluid, and the process repeated. Hardening may also be effected by injecting the chromic acid fluid into the vessels.

Müller's fluid for hardening consists of $2\frac{1}{2}$ parts of bichromate of potash, 1 part of sulphate of soda, and 100 parts of distilled water. The tissues in small pieces are to be soaked for a month or longer.

In Germany, most observers transfer the specimen from the chromic acid solution to methylated spirit, or even to strong alcohol. The method of preparation pursued by me is very different, and depends upon all the processes of hardening, &c., being conducted upon tissues impregnated with glycerine. The chromic acid and bichromate of potash, like all other hardening substances and chemical reagents, may be dissolved in glycerine, which by me is used as *a general medium, and constitutes the basis of all my solutions, instead of water.*

By mixing together certain solutions, having opposite properties, compound fluids may be obtained, which will exert different effects, upon tissues according to the proportion of the different constituents present. A mixture of alcohol and acetic acid, renders sections of the spinal cord and nerves beautifully transparent, and by its aid many new points in minute structure have been demonstrated, which, as far as is known, can be demonstrated by no other process. This solution was employed by Mr. Lockhart Clarke in his investigations on the spinal cord. Mr. Lockhart Clarke has since recommended chromic acid (1 part), and bichromate of potash (2 parts), and water (1,000 parts), for hardening the cerebellum, and has introduced othe modifications. It was only after a very laborious course of investigation and repeated trials of every kind of admixture which he thought likely to produce the desired end, that Mr. Clarke hit upon that most useful fluid, alcohol and acetic acid. In his very first paper, before he had carried his observations upon the anatomy of the cord to any very great

extent, he described minutely the manner in which his specimens had been prepared, and thus liberally gave his fellow-labourers the advantage of carrying on investigations in this wide field of enquiry, at a time when he himself had only commenced his researches. Dr. Lenhossek, of Vienna, has adopted Mr. Lockhart Clarke's plan, and has made some beautiful specimens, which he exhibited in London some years since, and which were afterwards purchased for the Museum of the Royal College of Surgeons.

The solution of acetic acid and alcohol used by Mr. Clarke consists of one part of the former and three of the latter. No better example can be adduced of the great value of studying the chemical and physical characters of the tissues, and endeavouring to overcome, by particular methods of investigation, the impediments which exist to the successful demonstration of the anatomy of many structures, than is afforded by the use of this fluid. The proportions may be varied according to the properties which the new fluid is required to have. Mr. Clarke soon adopted a modification of his original plan, and was kind enough to send me, in 1866, the following directions, which I print in his own words :—"The spinal cord and medulla oblongata of man and the higher mammalia are to be cut into pieces of half or three-quarters of an inch long, and steeped in a solution of one part of chromic acid in 200 parts of water, for three weeks or a month. It is then preserved for use in a solution of about one part of *bichromate of potash* in 200 parts of water. For hardening the convolutions of the cerebrum and cerebellum, the solution of chromic acid must be weaker than for the spinal cord or medulla oblongata, that is the proportion of one part of the acid to four, or even five hundred parts of water ; but the portions of brain must be small, not more than half an inch thick, otherwise they become rotten before the acid has reached their centres. A little spirit added to the solution for two or three days, after the first day, will prevent this. The pure solution can be renewed.

"Spirit of wine is used to wet the knife or razor in making sections, which should be washed in water before they are placed in the solution of carmine. When sufficiently coloured, the sections are again washed in water, and placed for ten minutes or a quarter of an hour in strong spirit : after which, if they be thin, they are floated on the surface of spirit of turpentine, where they remain until they are quite, or nearly transparent, three or four hours, when they are removed to glass slides, on which a little Canada balsam has been previously dropped. If now examined under the microscope, they frequently show but little traces of either cells or fibres—a circumstance which seems to have caused Schroeder Van der Kolk, and some others, to abandon the method *at first*,—but if the sections be set aside for a little while, and treated occasionally with a little turpentine, the cells and fibres re-

appear, and present a beautiful appearance. Before they are finally covered with thin glass, they should be examined at intervals under the microscope, to see whether all the details of structure have come out *clearly;* and if so, as much Canada balsam must be used as suffices for mounting. If the sections be of considerable *thickness*, it will be found best to place them in a shallow vessel, the bottom of which is kept simply wet with turpentine, which can therefore ascend through them from below, while the spirit evaporates from their *upper* surface, for the *principle* of the method is this:—to replace the spirit by turpentine, and this by Canada balsam, *without drying* the sections. The method at first is attended with some difficulty, and practice is necessary to ensure complete success. Experience, also, may suggest, according to circumstances, certain modifications of the *exact* process here given." See p. 57.

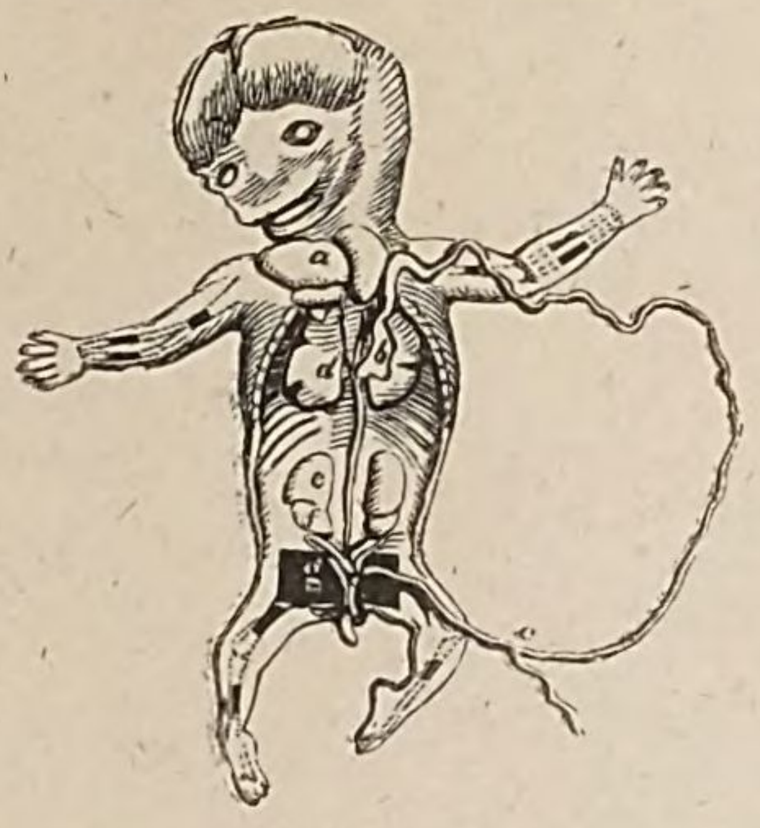
Clarke's process has since been modified by many observers. Instead of turpentine, oil of cloves or creosote has been used, and with great advantage. Chloroform has also been advocated, and the specimen afterwards immersed in a solution of Canada balsam in ether or chloroform. Dr. Bastian speaks highly of benzine, used for cleaning gloves, in place of chloroform.

By the addition of a few drops of dilute nitric acid to alcohol, a fluid may be obtained which has been found very useful in investigations on the different forms of epithelial cells. Another of these fluids was composed of the following ingredients; but of course useful modifications will be made by every practical observer, according to the properties which he desires the fluid should possess.

Water	1 ounce.
Glycerine	1 „
Spirit	2 ounces.
Acetic acid	2 drachms.
Hydrochloric acid..	½ drachm.

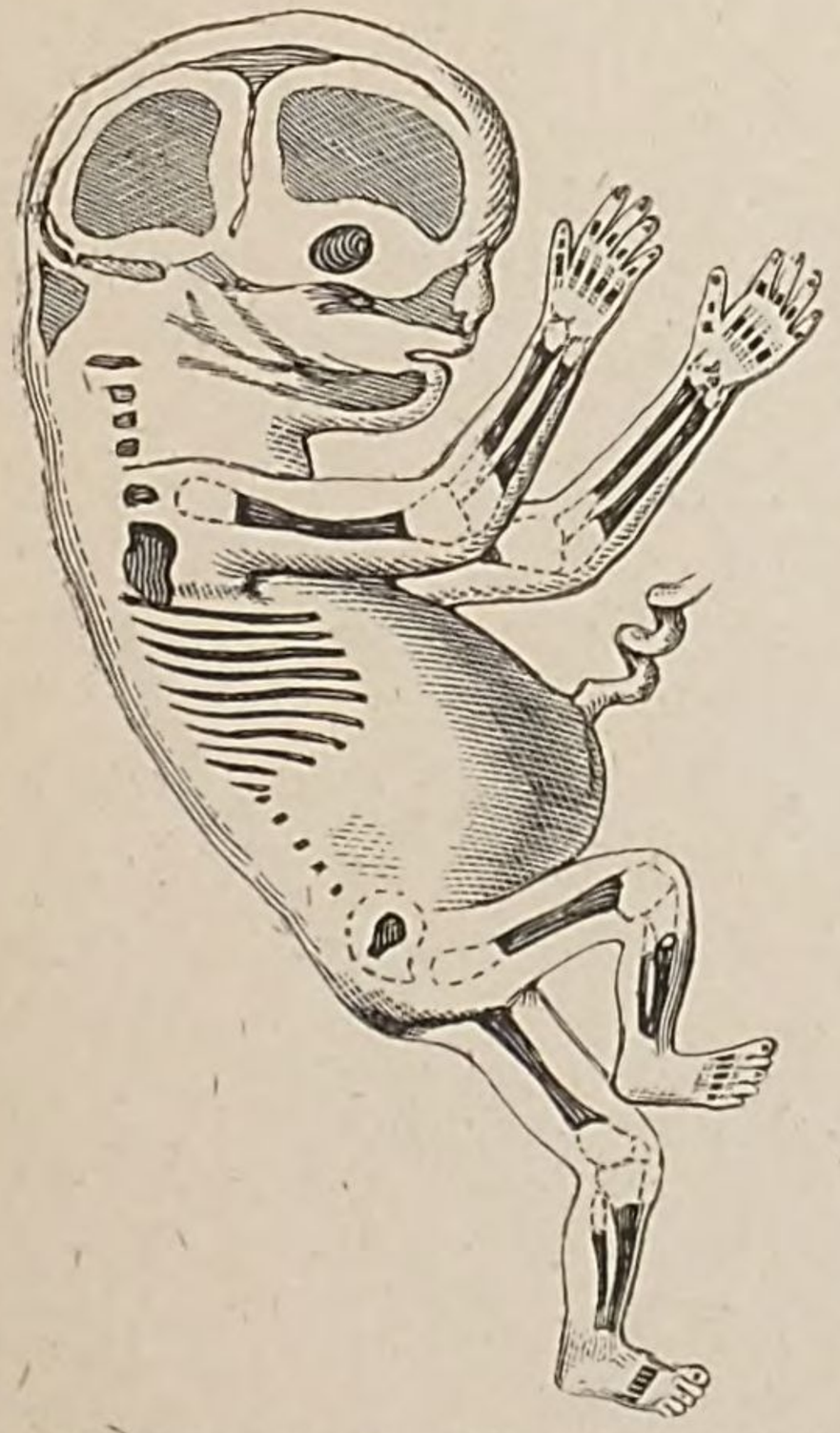
93. Alcohol and Soda.—In many investigations I have obtained excellent results from the use of a fluid composed of alcohol and solution of caustic soda, in the proportion of eight or ten drops to each ounce of alcohol. Many tissues are, at the same time, rendered very hard and transparent in such a mixture, and it is particularly adapted for investigations upon the character of calcareous matter deposited in tissues in various morbid processes. It is especially useful in tracing the stages of ossification in the early embryo. It renders all the soft tissues perfectly transparent, but exerts no action on the earthy matter of the developing bone. The most minute ossific points can therefore be very readily discovered. A fœtus, prepared by being soaked for a few days in this fluid and preserved in weak spirit, forms a very beautiful

Fig. 1.



Human Fœtus, about the eighth or ninth week of intrauterine life, soaked in alcohol and soda, and preserved in glycerine. *a*, heart. *b*, stomach. *c*, intestine, not yet much longer than the body The branch below the letter is the remains of the omphalo-mesenteric duct. *d*, lungs. *e*, supra-renal capsules. *f*, kidneys. *g*, remains of Wolffian bodies, with ovaries and genital ducts. Points of ossification are observed in the humerus, radius, ulna, last phalanges of the fingers, femur, tibia, and ribs. The ossification of the clavicle is advanced, but no ossific points are yet to be detected in the feet. Natural size. p 48.

Fig. 2.



Human Fœtus, about the eleventh or twelfth week. Ossific points are observed of considerable size. But one point exists in the os innominatum and two are seen in the scapula. The shading in the head and face indicates the formation of bone. The ossification of the first and third phalanges of the fingers and metacarpal bones has advanced, but at present there is only one point of ossific deposit in the tip of the great toe and one for the middle toe. In both drawings the development of the anterior extremities is much more advanced than that of the legs. Soaked in soda and alcohol for a few days, and preserved in spirit, Not changed since 1853—4. Natural size. p. 48.

[To face page 48.

preparation. A drawing of one, about the end of the second month, is given in pl. V, fig. 1, and in fig. 2, one about the end of the third month is represented. The first was prepared in 1853, and still preserves its transparency thoroughly. The practical advantages of such a plan over the usual very laborious process of dissection, in investigating the process of ossification in various bones, are obvious. This fluid will be found very useful in investigations upon soft granular organs. I also found it of special service when working at the anatomy of the liver.

Dr. Rutherford gives the following directions for the preparation of tissues previous to the commencement of his course on Practical Histology. As they will be found especially useful for the private student, I append them. "A month before the course begins, it is necessary to prepare some tissues, *e.g.*, to soften bone, to harden nerve textures, etc. A cat is a convenient animal to take. Let it be killed during digestion, open the stomach and wash out the interior with salt solution; send a stream of salt solution into the intestine; inject one-quarter per cent. chromic acid solution into the trachea, and distend the lungs (previously collapsed by opening the chest); tie the trachea, and place the distended lungs, together with the tongue, half of the stomach, a portion of the large and small intestines (cut into pieces an inch or so long), half of the liver (freely incised to permit the fluid to enter), the spleen, and some thin muscles from the limbs or abdomen, in a vessel containing a quantity of a quarter per cent. chromic acid solution sufficient to cover them. At the end of twenty-four hours, pour away the fluid, and replace it by a *large* quantity of fresh chromic acid solution of the same strength. Place the brain and spinal cord in methylated spirit for twenty-four hours, and then place the spinal cord, cut into small pieces, in a quarter per cent. chromic acid solution, and the brain, either whole or cut into small pieces, in chromic acid 1 part, potassium bichromate 2 parts, water 1200 parts. Procure a portion of human cerebrum (vertex), as fresh as possible, and treat it in the same manner. At the end of twenty-four hours, replace the chromic fluid on the brain and spinal cord by a large quantity of fresh fluid, and at the end of ten days place the *brains* in a fluid double the strength of that just mentioned. In the sclerotic of one eye make a number of small incisions, and place it in the chromic fluid used for the brain. Divide the other eye transversely about its middle, and place it in Müller's fluid (p. 46). Place the Schneiderian membrane in the chromic and bichromate fluid; the mesenteric glands, salivary glands, pancreas, ovaries, and uterus, and also the testes from another cat or a dog, in methylated spirit. Leave the periosteum on the bones, and place some of the limbs, together with the lower jaw and its teeth, in the fluid adapted for softening them.

"When all the tissues are hard, or nearly hard enough, and when the bones and teeth are sufficiently softened, place them in methylated spirit until sections are to be made. The spirit usually suffices to complete the hardening of chromic acid things. Chromic acid requires from one to six weeks to harden tissues. It is important not to leave them too long exposed to its influence, otherwise they are very difficult to stain with carmine and they may become brittle and useless for the purpose of section. Place a piece of intestine or stomach in Müller's fluid, in order that the connective tissue may be softened, and separation of the non-striped muscle permitted. Freeze a pig's kidney and make thin sections eight or ten days before they are wanted. Take care to place the kidney in the section machine so that the Malpighian pyramids may be cut vertically, and macerate the sections in Müller's fluid until the tubules separate readily (eight or ten days).

"In order to prepare the cochlea, kill a young guinea-pig, sever the head from the body, disarticulate the lower jaw, open the tympanic bullæ, remove the cochleæ and place them in a large quantity of a quarter per cent. chromic acid solution. The fluid must be changed every second or third day for a fortnight or three weeks. The cochlea of an old animal requires the addition of one-half to one per cent. nitric acid (Pritchard) to the chromic acid fluid at the end of four or five weeks. Let the cochlea remain in a large quantity of this doubly acidulated fluid until it is soft, then transfer it to methylated spirit until sections are to be made.

"Place a dozen fresh eggs under a hen or in a hatching-oven. Remove some eggs at the end of the twelfth, others at the twenty-fourth, others at the forty-eighth, and others at the seventy-second hour. Open the eggs very carefully under salt solution, cut out the embryos, and place them for two or three days in the chromic and bichromate fluid employed for the brain. Then transfer them to methylated spirit, and finally to absolute alcohol to complete the hardening."—"Notes on the course of Practical Histology, given in King's College, 1873."

CHAPTER III.

On Preserving Specimens Permanently.—Dry Preparations.—Specimens preserved in Fluids.—Alcohol.—Glycerine.—Glycerine Jelly.—Naphtha and Creosote.—Carbolic Acid.—Chloride of Calcium.—Acetate of Potash.—Chromic Acid.—On applying the thin Glass Cover, and of preventing the inclusion of air bubbles.—Of Preserving specimens in Canada Balsam, and Gum Damar.

To be permanently preserved, tissues must be (1) thoroughly dried and kept dry, or (2) they must be immersed in some resinous substance or varnish which undergoes little or no change, or (3) they must be immersed in some fluid medium which is miscible with water and aqueous fluids, but which has the property of preventing decomposition, and will not of itself become decomposed.

The first method is of service only in the case of few objects, and is only applicable to anatomical and pathological specimens of the most general kind, which are to be subjected to examination under low magnifying powers only, such as hair, horn, nail, bone, teeth, and a few others. Objects to be mounted dry may be placed in a dry cell (H. to W., § 114) and carefully protected from the dust by being covered with a piece of thin glass. Almost all specimens of interest to the practitioner require however to be immersed in some transparent medium. The second plan is therefore of wider application, and by its aid some peculiarities of tissues can be clearly made out. From some specimens preserved in Canada balsam, gum damar, and other materials of the kind, certain important facts may be ascertained, which cannot be demonstrated in specimens mounted upon other principles.

But the last plan is the most satisfactory, and for the display of many of the minute structural characters of the moist tissues of man, animals, and vegetables is the only one known to us. In all the higher branches of microscopical research we have to work upon tissues in a moist state, which can only be preserved in media miscible with water. The preservative action of such media may be due either to a chemical property of coagulating albuminous fluids, or some other chemical action exerted upon the matter of the tissue itself, or it may depend upon the physical properties of the medium employed to replace the water, and thus, with the growth of bacteria and fungi, decomposition of the animal or vegetable matter may be prevented. Syrup and glycerine

in weak solutions readily undergo change. Low organisms will grow in them freely, and fungi will multiply and destroy the tissues preserved in such media. But strong solutions of these substances will keep for indefinite periods of time without change. To syrup there is however a fatal objection—the tendency to crystallisation at a low temperature. Chloride of calcium, acetate of potash, and strong solutions of other deliquescent salts are preferred by some observers for the preservation of many animal and vegetable tissues, but in my hands glycerine has proved the most advantageous, and most generally applicable of all the media I have tried, and I have not found it advisable to use any other fluid for the preservation of specimens. As my conclusions upon this matter are however different from those of other observers, I shall describe in this and other books the methods recommended by others as well as those which have proved most satisfactory to me, and which, at least in my hands, have been very successful. It will be more convenient to describe the second method of preserving tissues (§ 102) after the third plan referred to has been considered.

94. Alcohol.—Formerly, tissues used to be preserved in alcohol, but for modern microscopic research alcohol is almost useless. Still it is recommended by almost every author. The spirit now used is methylated spirit which may be purchased at the rate of about 5*s*. a gallon. It may be diluted with an equal bulk or twice its bulk of water, but it so modifies delicate structure as to be inapplicable for specimens which are to be examined by high powers.

95. Glycerine.—As I have already stated in page 31, glycerine is a very useful medium for the examination of specimens. It is also a most valuable preservative fluid, and if certain precautions be taken is adapted for the preservation of all tissues, from the most delicate transparent tissues of an animalcule or jelly fish to the hardest and densest textures. In Chapter VII I shall describe in detail the different ways in which glycerine may be used. The strength may be increased at pleasure, and other fluids and the various test solutions may be added to it as may be desired. The glycerine now used is Price's patent glycerine, which may be purchased for 3*s*. a pound. If weak solutions of glycerine are required, the growth of fungi must be prevented by adding to each ounce of the fluid three drops of carbolic acid, or a fragment of camphor. The most curious and fanciful objections have been raised to the use of this substance, and the most extraordinary discoveries have been made by those who have used it a few times, and have considered it necessary to publish to the world the results of their "experience." Some have found that tissues shrink up in such a manner that nothing is to be made out, while others bear most positive testimony to the fact that connective tissue swells out in glycerine to such an extent as to have led some to believe that the appearance was due to a re-

markable pathological change. All forms of fibrous and connective tissue may be mounted in glycerine without shrinking on the one hand or swelling out on the other. Any one who studies the numerous methods of preparing tissues which are being continually invented and modified, and forced upon the attention of students, will find that, after all, glycerine is getting into increased favour, and some of those who have expressed themselves against its use have found glycerine of great service. For instance, Dr. Klein used to mount his sections of cornea, prepared by the gold process in Canada balsam or damar, but, in a paper read before the Microscopical Society, March 6th, 1872, he recommends that the sections of the cornea, after remaining from one to two hours in a half per cent. solution of chloride of gold, should be "washed in distilled water, and exposed to the light in distilled water for from twenty-four to thirty-six hours (the water being changed twice, or oftener). *The cornea is then placed in glycerine and water* (glycerine one part, water two parts)."—"Monthly Journal Mic. Science," April, 1872, p. 157. Dr. Klein speaks of the "great advantage" of the new method over the old one, and says that the nerve fibres are very sharply defined. It is much to be regretted that this plan had not been adopted before. I am quite sure that if Dr. Klein had had more experience in the use of this medium, many of the condemnatory remarks offered by him with respect to some of my observations would not have been published.—See "Monthly Microscopical Journal."

96. Glycerine Jelly is a very useful medium for preserving anatomical specimens. The mixture may be made as follows:—A certain quantity of gelatine or isinglass is allowed to soak for some time in cold water, until it swells up and becomes soft. It is then placed in a glass vessel and melted by the heat of warm water. It may be clarified, if necessary, by first adding to the cold gelatine a little white of egg, then boiling the mixture and filtering through fine flannel. To this fluid an equal quantity of strong glycerine is added and well mixed with it. This mixture may be kept for any length of time, and a very slight heat is sufficient to render it perfectly fluid. Mr. Rimmington, operative chemist, Bradford, prepares some very clear and transparent glycerine jelly. This may be obtained in small bottles free by post for 1*s.* 4*d.*

Gum and glycerine is also a very useful preservative medium.

97. Naphtha and Creosote in water form a preservative fluid, useful for keeping some animal tissues which are required to retain the same character they exhibited when immersed in water.

Naphtha and Creosote Fluid.

Creosote	...	...	3 drachms.
Wood Naphtha	...	...	6 ounces.
Distilled Water	...	...	64 ounces.
Chalk, as much as may be necessary.			

The naphtha and creosote are first mixed together, and to the mixture as much prepared chalk as is sufficient to make a smooth thick paste, is to be added. Then pour in gradually the water, mixing it with the other ingredients in a mortar. Add two or three small lumps of camphor, and allow the whole to stand for a fortnight or three weeks, occasionally stirring it. The almost clear supernatant fluid is then poured off ready for use.

98. Carbolic Acid.—The preservative qualities of this substance are now well known. A solution for preserving and mounting tissues may be made by adding to 100 parts of distilled water, 1 part of carbolic acid. Both animal and vegetable tissues may be preserved in this medium. Carbolic acid water is, I think, likely to prove very valuable, and has been recommended by many observers. It will probably supersede the creosote fluid. In using these watery solutions it is necessary to be careful that the specimen is thoroughly impregnated with the preservative fluid before it is mounted permanently. It should soak for two or three days before being put up.

99. Chloride of Calcium.—For preserving many vegetable tissues as well as specimens of bone, hair, teeth, and other hard substances, a saturated solution of chloride of calcium has been recommended.

Acetate of Potash.—A saturated solution is made with distilled water. It has been highly recommended by many observers, but I am not aware of its special advantages over glycerine.

Acetate of Alumina has also been recommended by some observers; but granules often form in these saline solutions after a time.

100. Chromic Acid.—A solution of chromic acid is prepared by dissolving sufficient of the crystallised acid to render the liquid of a pale straw colour. A quarter per cent. solution answers best. It preserves structures exceedingly well, but renders tissues granular. It is particularly adapted for hardening the brain and spinal cord, but is also very useful for hardening many of the softer animal tissues. Tissues should be soaked in the fluid for twelve hours, and then transferred to fresh fluid, which should again be changed after a few days. (See also page 46.)

Upon the whole I am of opinion that the strongest glycerine and glycerine jelly are the most advantageous media for preserving animal tissues, and carbolic acid water and creosote fluid may be used for the preservation of various specimens, for which a fluid possessing the highly refracting properties of glycerine is not suitable.

I believe that our knowledge concerning the distribution of nerves and the ultimate arrangement of the secreting portions of various glands, the relation of bioplasm to the capillaries and nerve fibres, and other most important but delicate points of anatomical enquiry will be rapidly advanced, if students will act upon the principles laid down in

Chapter VII, and follow the directions there given, modifying the details as experiment and observation show to be desirable.

101. On applying the thin Glass Cover and cementing it in its place.—As specimens mounted in gum damar and Canada balsam require no cementing, the remarks in this section apply not to them, but only to preparations mounted in aqueous fluids.—In H. to W. I have described how to make cells for the preservation of microscopical specimens. Most practitioners will purchase the cells they require of the instrument makers, so that it is only necessary to give directions for placing the specimen in the cell and applying the thin glass cover. The preparation having been soaked for many days in the fluid in which it is to be mounted, sufficient of the latter is to be transferred to the cell to rise a little above the surface of the rim. Next the specimen is introduced and placed in the proper position in the cell. The thin glass cover, having been breathed upon, is to be allowed to fall gradually on the surface of the fluid, the superfluity of which may now be removed by blotting paper until the thin glass cover comes into contact with the upper edge of the cell. A little time is allowed for the edges to dry, or if glycerine is the fluid used, as much as possible of the excess is to be soaked up with blotting-paper. Very thin sections, or minute snips of morbid growth, do not require even a thin cell, but may be placed with the fluid on one of the glass slides. The undue pressure of the thin glass cover is prevented by the insertion of three small pieces of writing-paper or thin cardboard, about the one-tenth of an inch square. The excess of fluid is removed after the thin glass cover has been applied, and the slide near the thin glass, if wet, carefully wiped. In the case of very thin sections of healthy and morbid tissues, not even paper between the thin glass and the glass slide is necessary, but the specimen with the drop of fluid is placed on the slide and the thin glass applied with the same precautions, the greatest care being taken, as in all other cases, not to include any air bubbles.

Now, as to the cementing. The preservation of the specimens depends entirely upon this part of the operation of mounting being properly conducted. Many cements have been recommended, and it is extraordinary how frequently, by different observers, the very same cement and the very same preservative medium has been highly praised and seriously condemned. As the student will probably conclude, everything really depends upon the *manner* in which the mounting has been conducted. Some say, "never use Brunswick black," but I have specimens in fluid which have been perfectly preserved for twenty years, in cells the covers of which have been cemented with Brunswick black. At the same time, gold size, shellac in alcohol, a mixture of gold size and the last solution, and gum damar, are, in my opinion, preferable.

Dr. J. G. Hunt, of Philadelphia, gives the following receipt for a

cement for cementing the thin glass to the slide: -A solution of gum damar in benzole is to be made so thick that it will just drop readily. Finely powdered dry oxide of zinc is to be triturated in a mortar with a little benzole, and added to the damar solution in sufficient quantity to make it white when thoroughly stirred. This may be used as other cements (quoted by Dr. H. C. Wood, Junr., in his "Contribution to the Natural History of the Fresh Water Algæ of America."—Smithsonian Institution, Washington, 1873). This cement may be used for making cells, and even a deep cell may be made by painting successive layers one over the other, each being allowed to dry thoroughly before a new one is applied. As a last coating, there is no objection to a thin layer of sealing-wax varnish (sealing wax dissolved in methylated spirit).

Other cements are Canada balsam, dissolved in ether, chloroform, or benzole. The damar cement may be made by dissolving an ounce of damar in about two ounces or less of benzole. This cement may, however, be purchased ready prepared. Damar cements answer well for glycerine preparations. Specimens mounted in glycerine jelly require a layer of cement round the edge, to prevent the jelly from drying up.

In all cases, only a very thin layer of the cement should be applied first. This is left for some time to harden, and then another layer is painted on—a number of thin layers being preferable to a single thick one. If the cementing is properly conducted, the preparation will last for many years in a climate like England, but specimens exposed alternately to extreme heat and cold are almost sure to be lost in a few years. In all cases, preparations mounted in fluid must be kept flat in the drawers of the cabinet, and every preparation in the collection should be carefully examined at least once a year, and an additional layer of cement applied to any that may require it. If an air bubble should have got into the specimen, it may be easily removed and a little fresh glycerine introduced in its place, if that be the preservative fluid employed. In many cases it will, however, be requisite to remount the specimen entirely.

When a specimen is to be preserved or remounted in any fluid medium, it must always be allowed to soak in it for a considerable time before it is mounted. The best plan is to place the specimen in a few drops of the preservative solution in a watch-glass, protected from dust by a glass shade (pl. VIII, fig. 1), for two or three days before they are to be permanently mounted.

102. Canada Balsam has been more used than any other preservative medium, but of late solution of gum damar has been used in preference. Some objects may be dried and mounted in Canada balsam, but this is not satisfactory. A plan is described below by which objects may be mounted in the medium without being dried in the first instance.

On mounting Moist Tissues in Canada Balsam.—Moist tissues may be mounted in Canada balsam without being previously dried, by the use of strong alcohol, and then oil of cloves, or creosote. Rindfleisch employed the first. As is well known, Canada balsam will not permeate a tissue moistened with water; but the water may be removed by soaking in alcohol or in an alcoholic solution of acetic acid or soda, which does not alter the albuminous materials. When well saturated, the alcohol or the alcoholic solution, which now contains a little water derived from the specimen, may be changed for a little fresh alcohol, and after the specimen has been allowed to soak for some time in this, it may be removed to creosote or oil of cloves. The last solutions drive out the alcohol, and after the preparation has been placed once or twice in fresh portions of solution, it may be placed on a glass slide. The tissues may then be thoroughly impregnated with Canada balsam or damar solution, and mounted permanently. Thus, although neither Canada balsam nor damar possesses the property of wetting a tissue containing an aqueous fluid, it and similiar media may be made to permeate it in the manner above described. The process may be modified in various particulars, according to the particular tissue which is to be mounted. See also p. 46.

Thin sections of tissues, such as the brain or spinal cord, may also be moistened with a medium not miscible with water, by allowing them to lie for some time upon the surface of the fluid in a shallow dish. Another dish, containing a little strong sulphuric acid or some chloride of calcium, may be placed near, and the whole covered with a bell jar. The sulphuric acid or chloride of calcium gradually absorbs the water which is thus removed from the thin section, and the turpentine or other medium gradually permeates the tissue and takes its place, but it is seldom this plan succeeds perfectly.

Canada balsam and damar have this very great advantage over other media—that when once mounted, the specimen retains its character for years, and is probably as *permanent* as anything can be. But unfortunately the most important characters of the great majority of objects of interest to us are not retained in specimens mounted in balsam. Although many still mount their specimens in balsam or damar, and the plan finds special favour in Germany, I must venture to speak against it, for I think that many of the views now entertained concerning the structure of certain organs would never have originated if the specimens had not been mounted in Canada balsam. The most important anatomical peculiarities of most animal tissues are entirely destroyed by the process. It is impossible to form any idea of the relative position of delicate structures lying one over the other, for in consequence of the contraction which has taken place, fibres which really lie above or below one another, appear in the mounted specimen upon precisely the same plane, and

delicate lines as of nerve fibres which, in the moist specimens can be moved slightly over one another, and can be proved to be quite distinct and separate, and really pursue a tortuous course on many different planes, appear in balsam preparations to be fused together forming one fibre, and seem to run in perfectly straight lines. Capillaries shrink, and oftentimes vessels which are distinct from one another appear to be connected. But perhaps the greatest differences of opinion arising out of this plan of preparation are those which are now held with reference to the existence of very minute tubes connected with capillary vessels and lymphatics. The size injection employed seems to make slight rents in the capillaries, and to run into the spaces existing in various tissues, for example, in the narrow channels between the individual epithelial cells of cutaneous or mucous epithelial structures. The size slowly hardens in these tube-like spaces, and in the progress of drying contracts slightly, causing well-defined outlines to each cylindrical portion, and thus the appearance of minute *capillary tubes filled with injection, results.* Although such views have met with very warm support, they appear to me to rest upon a most unsatisfactory basis, and until I obtain stronger evidence from the preparations of others or from my own observations I feel compelled to dissent from the conclusions.

So with reference to the arrangement of the ducts of the liver. Even now, the existence of the finest ducts which I injected many years since is scarcely admitted, although a system of *far finer ducts* amongst the liver cells in every part of the lobule has met with general acceptance, and I believe that the inferences deduced have arisen almost entirely from the appearances seen in examining balsam preparations by observers who endeavour to make it appear that the objections they raise to the use of glycerine are as strong as, and more reasonable than, those which may be made to balsam. Upon paper this appears to be so, but when the two processes are practically compared, a very different conclusion is arrived at. The suggestion has certainly been repeated in Germany, by author after author, that the appearances described by me *may* have been artificially produced by the mode of preparation followed! Such observations only show that a most imperfect idea has been formed of what I have *actually seen* and have shown to others, in specimens prepared according to the plan of preparation advocated by me. Those who offer the opinion have either not tried the plan at all, or, having tried it have not succeeded, and have had no opportunity of seeing specimens prepared according to the principles laid down.

Another great objection to mounting specimens in balsam, damar, and such media, is that they are not susceptible of further dissection, while glycerine specimens can be removed after they have been mounted for years, and divided into several portions, dissected and remounted. By proceeding thus, I have been enabled, in many instances, to bring

out new points of importance, in old specimens. Not unfrequently it will be found that a change has gradually been induced which has rendered the minute dissection very easy, and has enabled me to separate the several constituent textures from one another, in a manner which would not have been possible when the preparation was first mounted.

CHAPTER IV.

Of Colouring Tissues Artificially.—Of Staining the Bioplasm of Cells and Tissues, and of Tinting the Formed Material.—Method of Colouring the Bioplasm, or Living or Germinal Matter.—The Carmine Fluid.—Of Tinting the Formed Material of Cells and Tissues.—Aniline Colours.—Thiersch's Fluids.—Nitrate of Silver.—Osmic Acid.—Solution of Chloride of Gold.—Dr. Klein's Directions for Staining the Nerves of the Cornea.

Of the many artificial processes proposed for the purpose of rendering a transparent and perhaps invisible substance more or less distinct, the process of staining is the most efficient. It is surprising how many important facts relating not only to the build or structure of the tissue, but concerning the mode of its formation, and the manner and rate of its growth, have been ascertained by adopting this process. And it is quite certain that what has been already discovered bears a very small proportion to that which yet remains to be ascertained. Not only may points of the greatest interest be made out in the structural arrangement of tissues, but the germinal or living matter, or bioplasm, which is alone concerned in tissue-production can in this way only be accurately demonstrated and distinguished from less important constituents of the tissues. In the early stages of disease, changes which would be inappreciable to the eye if the specimen were prepared by the ordinary methods are rendered obvious enough if the process of carmine staining described in this chapter is resorted to. The number of masses of living matter, or bioplasm, in all tissues is found to be enormously greater than has been hitherto supposed. In truth, only comparatively few of the bioplasts or masses of living matter (nuclei) of tissues are demonstrated by those observers who simply examine textures in water, serum, vitreous humour and the like.

The processes of staining are employed for two very different objects.

1. For colouring the *bioplasm*, or *living* or *germinal matter* of the cell or texture.
2. For demonstrating peculiarities in the build of the *formed material*, *cell wall*, *intercellular substance* or *tissue*, and for ascertaining the order in which the several parts of which it is composed have been laid down.

Since the year 1861, when I first described the great advantage of studying the arrangement of the bioplasm of the same tissues at different periods of development, I have been actively engaged in prosecuting further investigations and extending the generalizations then arrived at. Not only the methods of investigation, but the mode of description of the changes in tissues adopted by me have been followed out by other observers in this country, in America, and on the continent. And, although as must inevitably be the case in the times in which we live, trivial objections and the by no means disinterested opposition of opponents have had undue influence in raising doubts in the minds of students concerning the accuracy of the results and the correctness of the inferences,—the principles of investigation as well as the general conclusions advocated by me, find an increasing number of advocates.

I.—Of Colouring the Bioplasm.

103. Method of Colouring the Bioplasm, Germinal, or Living Matter.—As I have shown, *bioplasm*, or *living matter*, is in all cases *perfectly clear and transparent*. It never exhibits *structure*, and is *invariably colourless*. It possesses an acid reaction, or, to speak more correctly,—an acid reaction is always developed immediately after the death of any form of living matter. Hence, if any alkaline solution of colouring matter from which the colour is precipitated or fixed by an acid, be caused to pass into the bioplasm of a cell or tissue before decomposition has commenced, the alkali of the colouring fluid is neutralised by the acid there developed, and the colour is retained. It may be precipitated in a state of very minute subdivision, or may combine with some of the constituents of the recently dead bioplasm and form with them compounds insoluble in weak acids. The tissue itself, or formed material, being ordinarily bathed with an alkaline fluid does not take the colour, and hence, by carrying out the process with due care, the *bioplasm* or *living matter may be coloured while the formed material or tissue remains perfectly colourless*. Any one can satisfy himself of this fact by placing upon a glass slide a few liver cells from any animal immediately after its death. If a drop or two of the solution of carmine in ammonia (page 65) be allowed to flow over the cells, the nucleus and nucleoli which constitute the bioplasm, or living matter, of each cell will be distinctly coloured in the course of a few seconds, while the outer part of the so-called "cell," composed of *formed material* which has been traversed by the same fluid, will not be affected.

It has been objected that formed material can be coloured by carmine as weli as bioplasm, or living or germinal matter. This is true in a sense, for it need scarcely be said that dead and dry things, such as paper, cotton, hair, wool, and the like may be stained with carmine. White

fibrous tissue may be deeply dyed if a strong solution of carmine be employed, or the texture be left to soak for a considerable time. In like manner the axis cylinders of the nerves (which I believe to be formed material only), and yellow elastic tissue, nay, even bone and teeth, may be stained. But on the other hand, the nuclei or masses of bioplasm taking part in the formation of every one of these tissues may be permanently coloured, although the tissue itself which is bathed on all sides by the fluid, remains perfectly free from all colour. If, however, the formed material be first impregnated with a little weak acid and then placed in the carmine fluid, the carmine is retained as in the case of bioplasm. While if bioplasm be rendered *alkaline*, or allowed to become alkaline after death, it will not take the colour. The fact of real interest and importance is that the bioplasm of every living thing, when fresh and without previous preparation, is invariably coloured deeply, and in a short time; while *under the same circumstances*, its formed material, although in contact with the carmine fluid, is not coloured at all, and this—notwithstanding the fact that all the fluid that has reached the bioplasm has traversed this formed material in the first instance.

This important conclusion has been in no way disturbed, and may now be regarded as a general fact of great importance, and true of all things living and at every period of life. That many will fail in obtaining the results I have described, and which are illustrated by my specimens, will, I fear, have to be admitted, but that for such a reason my conclusions should be set down as untrustworthy, as some have tried to enforce, is curious enough. If proper precautions be taken, it will be found that the bioplasm of any tissue, vegetable as well as animal, at any period of life, may be beautifully coloured while any formed material through which the colouring solution has passed remains perfectly unaffected by the carmine fluid.

As long ago as 1858–59, I commenced to study the changes occurring in the same texture at different periods of its growth, with the aid of an alkaline carmine fluid of uniform strength, and repeated my observations upon many different tissues of man, the higher and lower animals, as well as upon the simplest and most complex vegetable organisms, and was led to the general conclusions published in my paper "On the structure of Tissues, with some observations on their growth, nutrition, and Decay" (Archives of Medicine, 1860). Since that time I have continued to employ the same plan of observation, improving it from time to time, as new suggestions occurred to me. More careful and prolonged observations upon the part of many other observers have strongly supported the general view I ventured to advance with regard to the structure and formation of all tissues,—namely, that every anatomical element or "cell" consists of matter in two distinct states or conditions of existence; A, *living, that is capable of growth and of changing the*

matter it takes up; and B, *formed, perhaps exhibiting structure, but passive, lifeless, incapable of growth, or of forming new structures or compounds.*

In all cases, by taking certain precautions, we can distinguish in any given tissue what part is living and growing, and what part has already been formed and has ceased to undergo *vital* changes. In considering the *structure* of any texture or in describing the alterations occurring in the course of disease, we may regard it as being made up of living germinal matter, or *bioplasm*, and *formed material.*

But bioplasm, or living matter, will take the colour long after the death of the animal, if decomposition, in which case an *alkaline* reaction would be developed, is prevented, as may be effected by alcohol and some other preservative fluids. So that specimens which have *been preserved for some time*, may be stained with the carmine solution, provided they were immersed in a preservative medium very soon after death; but the results will not be so satisfactory as if the tissue had been plunged into the carmine fluid when perfectly fresh. Peculiar and exceptional appearances may often be produced by the action of preservative media. By some the bioplasm is in part, or entirely, coagulated, and thus modifications are brought about which may lead the observer to draw very erroneous inferences. In practice, therefore, it is always desirable to immerse the texture when it is perfectly fresh, in fact, as soon as possible after it has been removed from the animal.

Many plants and lower animals may be killed by immersion in the carmine fluid, but some few resist it, and will even live in it for some time. In this case the staining will not occur until death is caused by exposure to heat, the action of alcohol, or in some other way.

The Rev. Lord S. G. Osborne, in June, 1856, showed that the nuclei of the cells of plants which were allowed to *grow* in a carmine solution, were more deeply tinged by carmine than other parts of the cell ("*Vegetable Cell Structure and its Formation, as seen in the early stages of the growth of the wheat plant.* '*Trans. Mic. Soc.*,' vol. v, plate iv, 1865"). Welcker and Gerlach subsequently employed the ammoniacal carmine solution for colouring nuclei. But previous to the delivery of my lectures to the Physiological Class of King's College, in the session of 1859–60, no one had pointed out the striking fact that by the aid of the alkaline carmine fluid, the matter directly concerned in growth, development, formation, and nutrition of all tissues of all beings in nature, could be distinguished from the tissue and other matters formed from and by this matter, which was shown to be the only part of the tissue or living being in a living state.

Gerlach was, I believe, the first to use a concentrated solution of carmine in ammonia, in which were placed specimens, for instance,—sections of brain or cord previously hardened by chromic acid, for from ten to fifteen minutes. They were then well washed in water for some

hours, and treated with acetic acid. The water and acid were removed by immersion in alcohol, and the sections afterwards mounted in Canada balsam. Gerlach afterwards found that dilute solutions (two or three drops of the ammoniacal solution of carmine to an ounce of water), and maceration for two or three days, afforded better results. In this process the specimen was hardened before it was stained.

Bioplasm may be coloured with Prussian blue fluid (p. 87), if it be rendered alkaline in the first instance by soaking the texture in a weak solution of ammonia. I have prepared some beautiful specimens as follows :—A weak solution of ammonia was injected into the vessels, and, after allowing twelve hours or more for the tissues to become thoroughly permeated, the finest Prussian blue fluid was introduced. The latter, in a short time, passed into the very substance of the bioplasm, which was tinged much more deeply than the surrounding substance. The liver cell may be thus impregnated with the blue in every part. It seems probable that by prosecuting more detailed enquiries in this direction, we might learn something concerning the physical arrangement of the matter constituting the formed material. Specimens prepared in this way enable us to prove the unsoundness of the old notion concerning the supposed cell wall and cell contents; but in endeavouring to draw correct inferences regarding the natural arrangement of the parts prepared in this way, it must not be forgotten that the alkaline ammonia may have effected alterations in the formed material, and modified its structure in an important manner.

As already stated, anything may be stained with the carmine fluid. If formed material or non-living material of any kind be slightly acidulated, and then transferred to the carmine fluid, the carmine will be deposited in its substance. Nerve fibres may be stained with carmine fluid. Even the white substance of Schwann may be deeply coloured. The colouring of the axis cylinders of the dark-bordered fibres and the processes of nerve cells is easily effected; but it must not be inferred from this fact that these structures consist of bioplasm. They are formed material, the former consisting of a very firm and unchanging form of formed material. As I have elsewhere stated, any form of fibrous tissue, nay, even bone and dentine may be deeply stained by carmine; but the bioplasm of these and all other textures may be coloured, while the formed material remains perfectly colourless, and this is the important fact to bear in mind in preparing tissues for observation.

The so-called "vacuoles" in growing tissues are proved by the carmine process to be occupied not with mere passive watery fluid of little or no importance, but with colourless bioplasm which can be satisfactorily demonstrated by this method of investigation, by which also its true nature is determined. So, too, from many free bioplasts, as

white blood corpuscles, lymph, and pus corpuscles, and many other bodies consisting chiefly of living matter, a spherical colourless body, like a minute bead of fluid serum, often proceeds after these have been for some time on the glass slide. In many instances, by the careful addition of a drop of the carmine fluid, I have been able to prove that this consisted of bioplasm, which, in fact, had separated from the part of the original mass that had undergone change, and had, in fact, passed into the lifeless state and mingled with the small portion of formed material which existed originally in connection with the living corpuscle.

104. The Carmine Fluid.—The following is the composition of the carmine fluid :—

Carmine, 10 grains.
Strong liquor ammoniæ, ½ drachm.
Price's glycerine, 2 ounces.
Distilled water, 2 ounces.
Alcohol, ½ ounce, or more.

The carmine in small fragments is to be placed in a test tube, and the ammonia added to it. By agitation, and with the aid of the heat of a spirit-lamp, the carmine is soon dissolved. The ammoniacal solution is to be boiled for a few seconds and then allowed to cool. After the lapse of an hour, much of the excess of ammonia will have escaped. The glycerine and water may then be added and the whole passed through a filter, or allowed to stand for some time, and the perfectly clear supernatant fluid poured off and kept for use. This solution will keep for any length of time in a well-stoppered bottle. If a little carmine is deposited, owing to the escape of ammonia, one or two drops of liquor ammoniæ may be added to the four ounces of carmine solution.

The carmine fluid I have found most useful for staining the bioplasm, the living or germinal matter of tissue (nucleoli nuclei, protoplasm), and which I now employ for all specimens, is the above, but it may be diluted in some instances with advantage. Mr. Atkinson tells me that he has found that for the cord it may be diluted with seven times its bulk of water. More alcohol is required in some cases, but a few experiments will soon enable the observer, engaged in investigation, to ascertain the degree of dilution most advantageous.

The rapidity with which the colouring of a tissue immersed in the carmine fluid takes place, depends partly upon the character of the tissue and partly upon the excess of ammonia present in the solution. If the solution be very alkaline the colouring takes place too quickly and will be too intense, and much of the soft *tissue* or imperfectly developed formed material around the bioplasm will be destroyed by the action of the alkali. If, on the other hand, the reaction of the solution

be neutral, the uniform staining of tissue and bioplasm may result, and the appearances from which so much of importance in connection with the growth and formation of tissue is to be learnt, will not be produced.

Some tissues absorb the colour very slowly. Fibrous tissue, bone, and cartilage, even in very thin sections, will require twelve hours, or even more ; but perfectly fresh, soft, embryonic tissues, very thin sections of the liver and kidney, and thin sections of morbid growths rich in bioplasm, may be coloured in half an hour, while the cells of the above structures, placed on a glass slide, may be coloured in less than a minute. I have often coloured the bioplasm of the fresh liver cell *in a few seconds*, by simply allowing the carmine fluid to flow once over the specimen.

When the vessels are injected with the Prussian blue fluid the carmine fluid requires to be sufficiently alkaline to neutralise the free acid present. The colour of the blue is temporarily lost, but is restored when the specimen is afterwards transferred to the acid glycerine. The permeating power of the carmine solution is easily increased by the addition of water or alcohol, or both, but difficulties of other kinds arise if the fluid be too dilute.

After the specimen has been properly stained, it is to be washed in a solution consisting of—

Strong glycerine, 2 parts.
Water, 1 part.

After the lapse of five or six hours, or more, it may be transferred to the following acid fluid :—

Strong glycerine, 1 ounce.
Strong acetic acid, 5 drops.

After having remained in this acid fluid for three or four days, it will be found that the portions of even soft pulpy textures have regained the volume they occupied when fresh—but it is often necessary to increase the strength of the glycerine more gradually than has been indicated in these directions. (See also Chapter VII.)

II.—Of Tinting the Formed Material.

105. Of Tinting Tissue or Formed Material.—The *formed material* differs much in consistence and properties in different cases, but it never manifests the properties or powers characteristic of the bioplasm. It is often so very transparent that it appears, even to the highest powers, as if it was perfectly structureless, but in many cases indications of structure, and in some, peculiar and remarkable structural arrangements may be discerned by the aid of tinting processes. Every observer is familiar with the assistance derived from allowing very

transparent textures to become partly covered with insoluble particles suspended in the fluid in which they are immersed. Any insoluble powder in a state of very minute division will often enable us to demonstrate an outline in the case of a tissue which cannot ordinarily be distinguished from the fluid which bathes it.

For the purpose of *tinting* a transparent texture almost any soluble colouring matter, which does not produce any chemical change upon the tissue may be employed. Some solutions, however, give much more satisfactory results than others. A dilute solution of the carmine fluid answers very well for many purposes, but I append several solutions which have been employed by different observers.

Coloured fluids have also been recommended for demonstrating cavities, spaces, or minute tubules existing in some textures. Not unfrequently a coloured fluid may be made to pass in the slight intervals existing between the epithelial cells of many textures, and the appearance is such as to have led many observers to entertain the opinion that there existed a system of very fine capillary tubes, much narrower than the finest capillaries, in epithelial and other textures. From the fact that the appearance in question is seen very commonly in injected specimens, it has been, I think, too hastily inferred that such apparent channels are real tubes connected with the vascular or lymphatic systems, and form a connecting network of tubes, by which free communication between the interior of the blood-vessels and lymphatics is established.

106. Anilin Colours.—The beautiful reds and blues which have been lately so largely used as dyes, popularly known in this country as mauve, magenta, solferino, have been much employed by microscopists. The colour is not very soluble in water, but is readily dissolved by alcohol. A grain of the colour, ten or fifteen drops of alcohol, and an ounce of distilled water, make a dark red solution; or the colour may be boiled in water, allowed to cool, and then filtered. This fluid colours *tissues* very readily. The coloured solutions known as Judson's dyes are in a very convenient form for the purpose of the microscopist, and can be purchased at most of the oilshops, for 6*d.* a bottle. They may be diluted with water or glycerine.

Magenta has been recommended by Dr. Roberts for showing a minute spot connected with the red blood corpuscles of man. ("On peculiar appearances exhibited by blood corpuscles under the influence of solutions of magenta and tannin."—"Proceedings of the Royal Society," vol. xii, p. 481, No. 55, April, 1863). The peculiar action exerted by magenta and tannin upon the red blood corpuscles (see below), has not yet been satisfactorily explained, but my friend, Dr. Hughes Bennett, of Edinburgh, tells me that, with the aid of very high powers, he has been able to demonstrate that the minute spot appearing

after the blood corpuscles have been soaked in magenta exhibits angles, and he considers that it is in fact a minute crystal which has formed upon the corpuscle.

Every kind of cell wall and delicate membrane may be coloured. The cilia of ciliated epithelium may be tinted while they continue to vibrate. As the substance of the cell becomes coloured, however, the action of the cilia ceases.

Thiersch recommends the following blue fluid, the composition of which I take from Frey :—Oxalic acid, 1 part ; distilled water, 22 parts ; indigo carmine, as much as the solution will take up. Another solution of oxalic acid and water in the same proportion is required. One volume of the first solution is mixed with two volumes of the last and nine of absolute alcohol. The mixture is then filtered, and is ready for use.

An anilin blue fluid may be thus made :—Soluble anilin blue, $\frac{1}{2}$ grain ; distilled water, 1 ounce ; alcohol, 25 drops. This fluid is not acted upon by dilute acids or alkalies.

Thiersch gives the following fluid for colouring textures of a lilac tint :—Borax, 4 parts ; distilled water, 56 parts—the borax is to be dissolved in the water, and one part of carmine added. The red solution is to be mixed with twice its volume of absolute alcohol and filtered. The precipitate of carmine and borax is redissolved in distilled water, and is ready for use.

Thiersch uses the following carmine fluid :—Carmine 1 part ; caustic ammonia, 1 part ; distilled water, 3 parts. Filter. One part of this solution is to be mixed with 8 parts of a solution of oxalic acid (1 part of the acid to 22 of water), and 12 parts of absolute alcohol. If the solution is orange coloured, instead of dark red, more ammonia is to be added, and the orange becomes red. The orange colour may also be used for staining. If crystals of oxalate of ammonia become formed, they must be separated by filtration.

Although *tannin* does not colour animal membrane, it alters its character to such an extent as to enable us to see many peculiar points of structure or arrangement not visible before, or it produces a chemical change upon the substance, from which we gain important information. The action of tannin upon the red blood corpuscle is very peculiar ; it has been specially studied by Dr. Roberts, of Manchester. The solution is made by dissolving two grains of tannin in an ounce of distilled water, and should be used within twelve hours after it has been made. One drop of blood may be mixed with four or five drops of the tannin solution and a portion of the mixture examined under the microscope.

107. Solutions of Nitrate of Silver.—Of late years nitrate of silver has been used for demonstrating minute and delicate pores, and for staining tissues. Recklinghausen and His have employed this plan with great success. A weak solution may be imbibed by delicate tubes,

and part being precipitated in the tube, perhaps as a chloride or in combination with some albuminous material, subsequently becomes decomposed by the action of the light, and a very dark line results. In this way the position of a previously perfectly invisible channel may be clearly demonstrated. Transparent connective tissue and the outer part of cells can thus be coloured, the bioplasm remaining perfectly colourless and transparent. The nuclei (bioplasm) by longer immersion will also be coloured. The explanation of this fact is probably as follows :—As long as the nuclei, or bioplasts, are alive they resist the action of the solution, but as soon as they are dead it is imbibed. The appearances may be made to vary very much by modifying the mode of procedure and the time which the preparation is allowed to remain in the solution. After soaking in the nitrate of silver solution for some time (from one minute to half-an-hour) the specimen may be placed in distilled water, or in a weak solution of common salt, in order to wash away the nitrate which adheres to the surface or occupies the intervals between the cells. When this has been effected, the specimen is exposed to daylight or sunlight until the requisite degree of blackening has been obtained. The strength of the solution employed may be varied according to circumstances. Recklinghausen uses a very dilute solution, consisting of 1 part of nitrate of silver to 400—800 of distilled water. I have used the nitrate of silver in solution in glycerine with advantage.

In staining thin membranes or thin sections of tissues, it will be found advantageous to pin them over a space cut out of a clean tablet of gutta-percha or hard wax, about an inch in length by about half-an-inch or more in width (p. 80). In this way, with the aid of very small pins, the membrane may be stretched while exposed to the action of the staining solution ; or the tissue may be stretched over a glass slide until it is stained. A quarter per cent. solution of nitrate of silver may be dropped upon the moist tissue, or the latter may be soaked in it in a shallow vessel. After being subjected, *in the dark*, to the action of the silver solution, the tissue is to be well washed in distilled water for twelve hours, and exposed to the light. The tissue may be mounted in glycerine.

The structure of the cornea has been investigated by His and others, after the tissue has been prepared with nitrate of silver solution. The so-called "intercellular substance" (formed material) only may be coloured, or, after the whole structure has been thoroughly impregnated with the solution, it may be soaked out of the formed material, while that taken up by the nuclei (bioplasm) is retained, and may be decomposed by being exposed to light. In this latter case the bioplasts appear very dark and surrounded by a pale brown formed material. His thinks that when the nuclei are coloured, the precipitate of chloride

of silver in the formed material is re-dissolved and absorbed by the nuclei, in which it is afterwards reduced by the action of the light.

108. Osmic Acid—(Os. O_2) has been strongly recommended for demonstrating delicate nerve structures by M. Schultze and Roudneff. Fat cells, oil globules, and white substance of Schwann and Myelin entering into the formation of various kinds of nerve fibres, become of a very dark colour and almost black. Other textures are neither coloured so quickly nor so intensely, and often exhibit only a brownish tint. So that by this substance nerve fibres ramifying in various textures may be stained, and thus distinguished from other elements of the tissue. The fat of adipose tissue is often deeply stained. Solutions of various strengths may be employed, from $\frac{1}{10}$ to 1 part of osmic acid in 100 of water. I have tried this plan, but have not gained anything by its use. Indeed, since 1858, I have been able to demonstrate finer nerve fibres without osmic acid than with the use of this reagent. These processes are capable of almost endless modification. It has been said that osmium preparations ought not to be mounted in glycerine, but in a saturated solution of acetate of potash. I have, however, an osmium preparation which has been in strong glycerine for five years, and still retains its characters.

109. Solutions of Chloride of Gold.—Weak solutions of percloride of gold have been much used of late years for colouring nerve fibres, which, after exposure to light, exhibit a blue or violet tinge. A solution containing from 2 to 1 per cent. in distilled water should be made. The tissue, after having been soaked for three or four minutes till it becomes straw-coloured, is to be well washed in distilled water, and then placed in very dilute acetic acid, containing 1 per cent. or less. The time for soaking in the gold solution varies with different tissues. From three to ten minutes is the usual time, but very thin tissues may be stained sufficiently in one minute. The tissue may then be washed and transferred to glycerine and exposed to the light, or it may be kept in distilled water for some hours, in the light, and mounted in Canada balsam or damar (see p. 56). The nerves become coloured in the course of a few hours. By this plan Cohnheim professes to have made out very fine nerve fibres, which, he says, pass from the plexuses in the cornea to intervals between the cells of the conjunctival epithelium, and, after reaching the surface of the structure, end in terminal free extremities. Many considerations make me think this view incorrect. I shall direct the reader's attention to the observations of Dr. Klein, who differs entirely from me, but goes beyond Cohnheim and others.

Dr. Klein has been very successful in preparing gold preparations, and the following remarks have been taken from his paper (Monthly Microscopical Journal, 1872). According to the method of Henoicque, the tissue treated with chloride of gold is placed in a vessel filled with

a concentrated solution of tartaric acid, and the whole is placed in hot water, when a rapid separation of the gold-salt occurs. Dr. Klein adopted Henoicque's plan, and obtained *always* and in every cornea, without difference of light, perfectly uniform and highly satisfactory results. I give the method almost in Dr. Klein's own words. From a rabbit just killed, Dr. Klein cut out the cornea with a narrow zone of the sclerotic, and placed it, after having removed with care the iris or ciliary body which was, in some cases, extracted with it, in a watch-glass with a half per cent. solution of chloride of gold, in such a manner that the convex surface of the cornea looked upwards, and the structure rested upon its sclerotic border. After three-quarters of an hour or an hour, it was removed from the gold solution and transferred, in the same position, to a vessel containing distilled water. Here it remained from six to ten hours in the light, when the colour of the cornea, which was at first yellow from the action of the chloride of gold, changed to a light grey or steel grey. The cornea so coloured is placed in a small glass flask with a wide neck, in which a small quantity (five to ten cub. cent.) of nearly concentrated filtered solution of tartaric acid has been poured. As soon as the cornea has imbibed this fluid and sunk to the bottom of the vessel, it will be remarked that its colour has become much deeper, being more or less of a greyish violet. The flask is now immersed in a capsule into which has been poured as much water at 40° to 50° C., as will reach the surface of the tartaric acid solution. After a very short time, often in a few minutes, the preparation assumes an intense violet-red colour, which continually increases until at last the cornea, when the water has quite cooled, appears of a dirty dark brown-red colour, and with a shiny velvety surface. It is now removed and washed for two hours or more either in common or in distilled water. From eight to twelve hours have now passed since the cornea was coloured with gold. After washing, the cornea is ready for examination. The epithelial layer, with a very thin layer of corneal tissue to which it adheres may be detached with the aid of forceps. It may be mounted in damar, or balsam, or preserved in glycerine. See p. 52.

In order to demonstrate the finest nerve-fibres of the frog's cornea, Dr. Klein recommends the following process. He says, "I pass a silk thread through the centre of the cornea of a healthy middle-sized or large specimen of *Rana esculenta*, and bringing it out again at the sclerotic ring, tie in it a loose knot to hold it fast; in short, I proceed in the same way as one does in inflammation studies. After the thread has remained from five to eight hours in the cornea, I cut out the latter with the greatest care, allow it to remain from a quarter to half an hour in pure half per cent. solution of gold chloride, and place it then in distilled water so long as the action of the light lasts, that is, until it has obtained the well-known dark violet-red or red-brown colour, a space of time

which varies according to the season from one to three days. Then I tear off from this the epithelium, together with a very thin layer of the corneal tissue, and enclose the remaining portion in glycerine."

The remarks made under the head of Nitrate of Silver, concerning the advantage of pinning out thin membranes, apply also to the gold process.

Many modifications of the above processes of investigation have been tried by me. I have found some advantage from using glycerine with the fluids, but at present I have no special plan to recommend. While I fully acknowledge the accuracy of many of the drawings and descriptions given of the appearances resulting from the use of nitrate of silver and chloride of gold, I am not convinced that many of the interpretations and conclusions which have been given and accepted concerning the structures demonstrated, are true. Some will, I think, have to be much modified in the future. The dark lines resulting from the silver process, which have been considered in many instances to be the outlines of epithelial cells, as for example in small vessels, mark, I believe, the lines of junction of the several elementary parts of which the tissue of the vessel consists. So, too, with reference to specimens prepared with gold, I am disposed to think that many of the lines which are rendered so very distinct by the black deposit will be found to have nothing to do with the transmission of nerve currents, and that many of the conclusions generally received will prove to have been too hastily adopted.

Violet Staining with Hæmatoxylin.—"The ordinary extract, hæmatoxylin, is rubbed down in a mortar with three times its bulk of alum, till both are reduced to a fine powder, and well mixed. A small quantity of distilled water may now be added, and the whole well rubbed together for 15 or 20 minutes. More water may now be added, and the solution, after filtration, should present a somewhat clear dark violet colour. 2 drachms of 75 per cent. alcohol may now be added to each ounce of the solution."—"Monthly Journal of Microscopical Science," December, 1872, p. 277.

Frey also prepares the above in this way. An aqueous solution of the extract of logwood is to be mixed with a solution of alum (1 part of the salt to 8 parts of water) till the deep red colour has become violet, and then filtered. These hæmatoxylin fluids may be used for fresh tissues, and for tissues hardened by chromic acid or alcohol. Tissues colour very rapidly and very deeply, in from half a minute to 10 or 15 minutes. They may be mounted in Canada balsam or damar.

CHAPTER V.

Of cutting thin sections of soft Tissues.—Instruments required.—Scalpels.—Double-edged Scalpel.—Valentin's Knife.—Of Machines for cutting thin Sections, or Microtomes.—Of the use of the Compressorium.—Of cutting thin sections of hard Tissues, such as Bone, Teeth, &c.—Of making minute dissections, and of dissecting under the surface Water, Spirit, or Glycerine.—Of magnifying Powers for Dissecting.—Of making Casts.

THERE is no more important operation in microscopical investigation than this. The student often requires thin sections of different textures, and whether he pursues the study of vegetable or animal physiology, or morbid anatomy, it is equally necessary to make a very thin section of the tissue which is to be examined; and upon the amount of skill he displays in cutting these sections, will mainly depend his success in investigation. The more deeply tinted, and the more complicated in its structural arrangement the tissue may be, the more important is it to obtain a section of extreme tenuity, for otherwise sufficient light cannot be transmitted through the section to enable the observer to see its structure clearly. The objects in a thick section, occupying different planes so much interfere with one another, that not one on any plane will be defined clearly, although the tissue itself may be tolerably transparent.

110. Of cutting thin Sections of Soft Tissues.—Sections of the large glands, and other soft tissues may be made with an ordinary knife, which should be very sharp. A clean surface is first cut, and then a thin slice is removed with a slow sawing movement of the knife, which is much facilitated by the application of a drop of water; indeed, whenever we require a very thin section of a soft tissue, the blade of the knife should always be well wetted with water or with the fluid in which the preparation is immersed.

The knife may be rendered very sharp just before cutting a thin section, by drawing the edge forwards first on one side and then on the other, upon a very smooth plate glass slide, or upon a smooth strop, or even upon a smooth piece of wood. In practice, however, I prefer the plate glass slide.

The most important instruments for making thin sections of soft

tissues are the following : scissors of different sizes, pl. VI, figs. 4, 5, 6 ; Valentin's knife, pl. VII, figs. 2 and 3 ; double-edged scalpels, pl. VII, fig. 1, or lancets mounted in handles, and a few other instruments, such as forceps, pl. VI, fig. 7 ; and needles of different sizes, pl. VI, fig. 3, mounted in handles, are often required in demonstrating minute structure. Tissues which have been hardened can be often cut into thin sections more readily by a sharp razor than by any other instrument. The observer should be provided with several razors, so that he may always have one or two sharp ones by him. Razors can now be purchased for 1*s.* each.

111. Instruments for cutting Thin Sections of soft Tissues.—*Double-edged Scalpel.*—For cutting very thin sections, a knife of the form represented in fig. 1, pl. VII, is very useful, and, where only sections of small dimensions are required, this will answer all the purposes of Valentin's knife. In cases, however, where a section is wanted of considerable extent, the latter instrument, or a section cutter, p. 75, must be used. The double-edged scalpel is made after the fashion of a common lancet ; it is not so wide, but should be quite as thin. When employed for making a section (after cutting a clean surface), the point is made to perforate the surface, and carried along at a proper depth, so as to cut its way out. The width of the section may then be increased by carrying the knife first to the right, and then to the left, until the desired width is obtained. Messrs. Weiss & Son, of the Strand, have made for me excellent knives of this kind.

Common Lancets mounted in handles will be convenient for cutting thin sections, but each side of the blade should be sharpened down to the point of insertion into the handle.

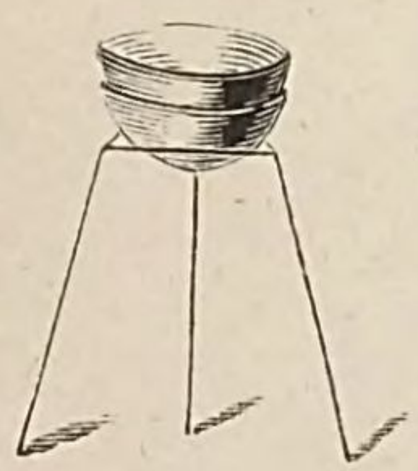
Scissors are also very useful instruments for cutting small thin sections of different tissues. The most convenient forms for this purpose are those represented in pl. VI, figs. 4, 5, 6. When thin sections of a tissue of no very great extent are required for examination, they will be removed with scissors more easily than with any other instrument.

Valentin's Knife has two blades, both perfectly flat on the opposed surfaces, very thin, and made perfectly sharp. By a mechanical arrangement, the blades may be easily separated from each other, or approximated to any required degree, according to the thickness of the section desired. The thin section is received between the blades, and is removed by separating them, and agitation in water. This instrument is of the greatest value in making thin sections of soft tissues of great extent, but it requires care to keep it in good order. It is very easily made blunt if used for cutting fibrous or cartilaginous textures. By its aid, most beautiful sections of the perfectly fresh kidney, liver, and other soft glandular organs may be obtained with the greatest facility.

Fig. 1.

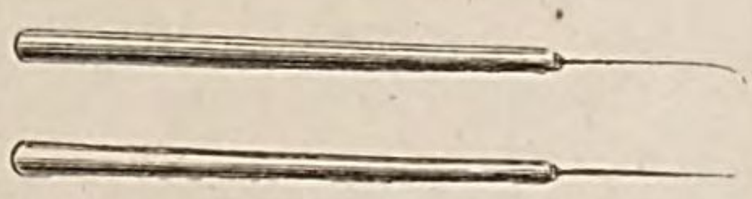
Spirit lamp, to which a wire stand is attached for supporting watch glasses, &c. p. 27.

Fig. 2.



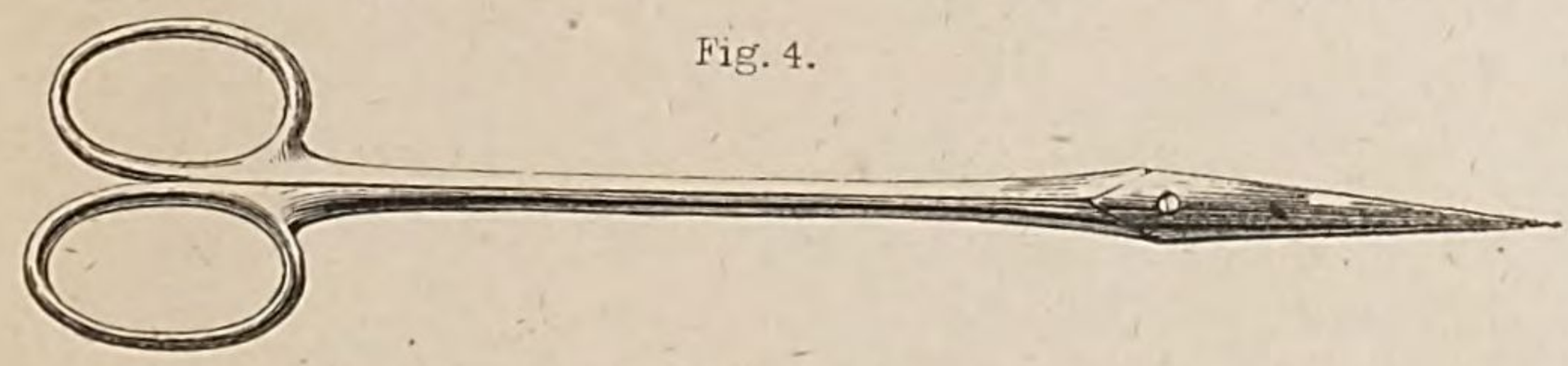
Porcelain basins, on tripod wire stand. pp. 27. 41.

Fig. 3.



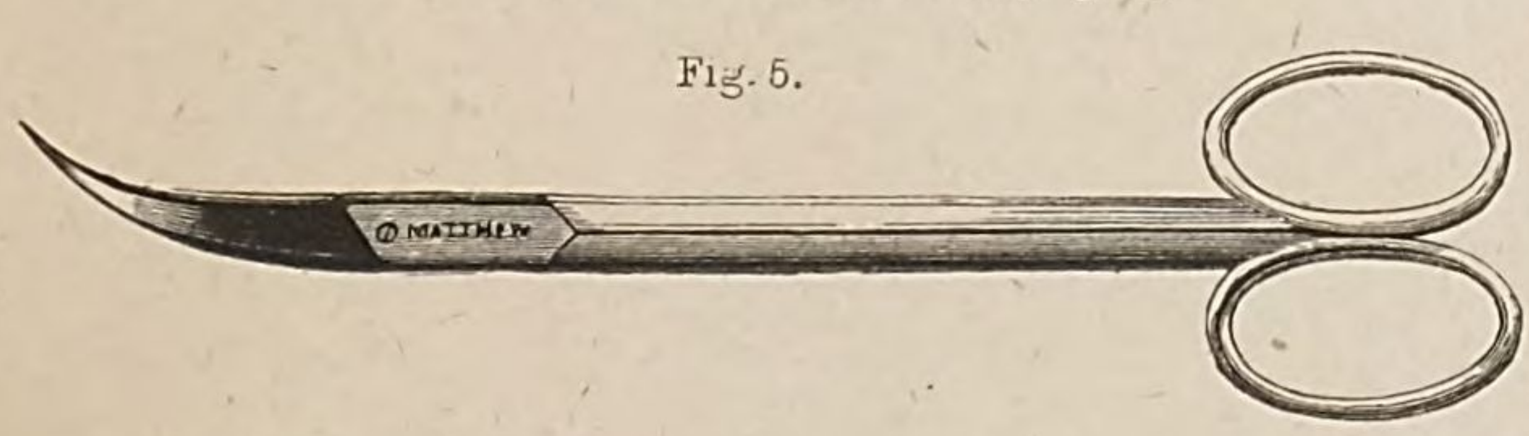
Needles, fixed in handles, for dissecting. pp. 27, 79.

Fig. 4.



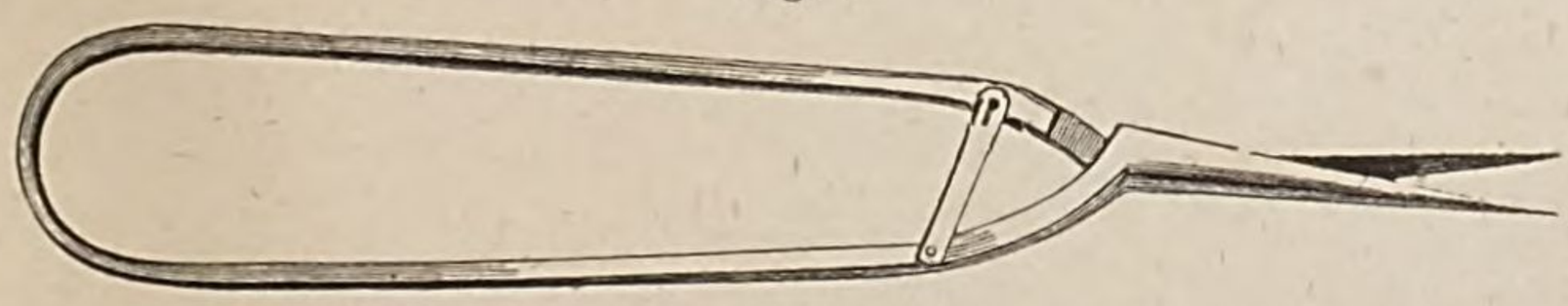
Fine straight scissors, for dissecting. p. 74.

Fig. 5.



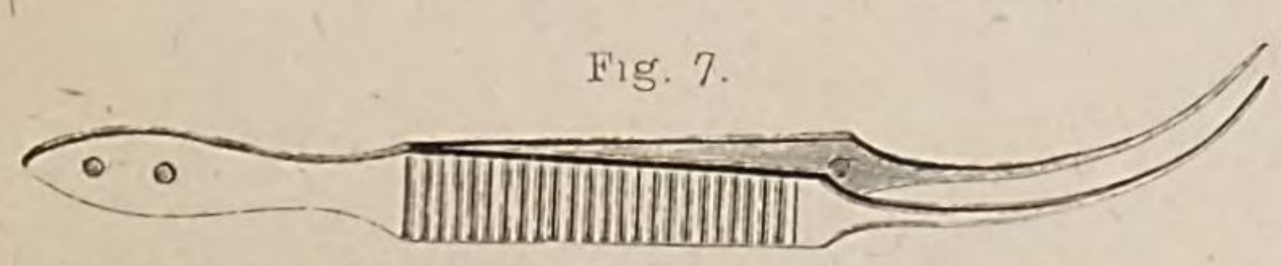
Curved scissors, for cutting thin sections of tissues. p. 74.

Fig. 6.



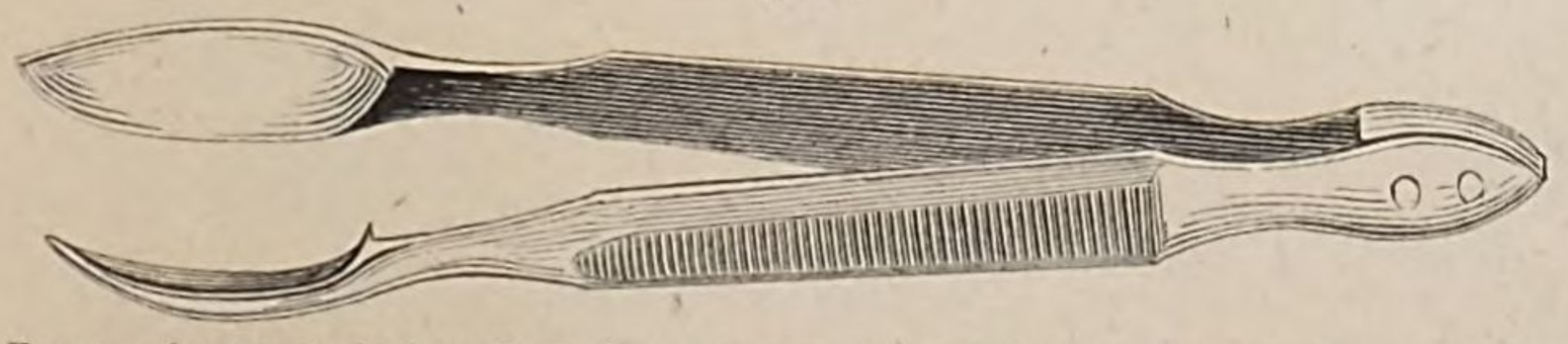
Small spring scissors, for making minute dissections. pp. 74, 80

Fig. 7.



Curved forceps, for making minute dissections. p. 47.

Fig. 8.



Forceps, for removal of small portions of viscid Sputum, for microscopical examination.

[To face page 74.

The blades should always be dipped in water, or in the fluid in which the specimen is immersed, just before use, for, if wet, the operation of cutting is much facilitated, and the section is more easily removed from between the blades. Immediately after use, the blades should be washed in water, and dried with a soft cloth or a piece of wash-leather. If a drop of water gets into the upper part of the knife where the blades meet, the screw must be taken out, and each blade cleaned separately. With care the knife may be kept in good working order for years.

Two forms of Valentin's knife are in common use; in one of these the blades are sharp on both edges, and of a lancet-shape, and in the other, which I much prefer, of the form represented in fig. 2, pl. VII. The best form of Valentin's knife, however, that I have made use of, is that represented in fig. 3, pl. VI, which was made by Mr. Matthews. The blades of this knife can be completely separated from each other and are easily cleaned. Moreover, the distance between the blades is regulated by a little screw, *a*, which is a most convenient arrangement. Mr. Matthews has lately much simplified the plan of making this instrument, thereby rendering it cheaper, without in any way impairing its usefulness. By adapting *two* screws to the blades he has been able to gain some advantages; not only can the blades be separated or approximated more readily, but greater firmness is obtained, and the perfect parallelism of the two blades is ensured. The knife may now be purchased for much less than formerly.

112. Of Machines for cutting Thin Sections. Section Cutters or Microtomes.—Although thin sections of tissues can be made without the use of any instrument except a good sharp knife, there is no doubt that where very thin sections of considerable extent are required other means must be employed, and for many years past, especially for making thin sections of the brain and cord, section cutters have been much used. Thin sections of fresh tissues may be cut after freezing (§ 86, p. 40), and Dr. Rutherford's section cutter, which will be presently described, has been so arranged that sections of fresh frozen tissues may be obtained.

Under ordinary circumstances before a thin section can be cut the tissue requires to be hardened. The best method adopted for this purpose is that of soaking for some time (from one to four weeks) in weak solution of chromic acid (pp. 46, 49). Many observers have used alcohol, and other media have been recommended. The tissue when properly hardened is removed to the section cutter, but before thin sections can be satisfactorily obtained, it is usually necessary to "bed" the hardened tissue in some medium that can be melted at a comparatively low temperature but becomes hard on cooling. Many *bedding preparations* have been suggested. Wax and oil has been used.

Dr. Rutherford recommends a mixture made of five parts of solid paraffin, which may be obtained in the form of paraffin candles, and one part of hog's lard.

The piece of tissue to be bedded may be freed from adhering moisture by blotting-paper, and then fixed in the centre of a small paper cup, corresponding in size to the opening of the section cutter, with the aid of a pin. The paraffin, or wax and oil, having been carefully melted in a water-bath is to be poured out and allowed to cool, when the paper may be torn away and the mass with the embedded tissue removed to the section cutter. For very delicate tissues Stricker recommends a strong solution of gum which may be hardened in alcohol after the tissue, also hardened in the same medium, has been immersed in it. The hardened gum with the contained tissue is then to be bedded in paraffin in the manner already described.

The original form of the section cutter in long use, for making sections of wood and other vegetable tissues has been described in "How to Work with the Microscope." But many improvements have been made in adapting this simple instrument to other purposes. Stirling's improved section cutter has been further improved by others. Dr. Rutherford has added an indicator, by which the thickness of the section can be estimated, as well as an arrangement for freezing tissues that are being operated upon. This instrument has been made by Mr. Hawksley, Blenheim Street, Bond Street, and latterly, by Mr. Baker, 244, High Holborn. Further improvements have been made as regards the table, some observers preferring steel to the ordinary smooth surface of brass, while Mr. Needham has had a microtome made with a plate glass surface. As regards the form of knife, an ordinary razor has been recommended. Dr. Matthews has improved the knife by having the shoulder ground down flush with the rest of the blade. It is, I think, better to have the knife or razor separate, and not to move on a hinge joint fixed to the table of the section cutter.

For researches on the structure of the brain and spinal cord, these section cutters are invaluable, and by the aid of sections cut by them most important facts connected with the course and distribution of nerve-fibres will unquestionably be determined. I am surprised that no one has yet traced the course of individual nerve-fibres, say in a convolution of the brain, or in a portion of the spinal cord, for it is certainly possible to do so at this time. From a well-hardened piece of brain or cord, numerous sections might be removed one after the other, and each one carefully mounted in the same position, and in the same manner. By examining these one after the other, it would be possible to trace many fibres in their course through a piece of nervous matter, as much as half-an-inch in thickness. With the aid of careful sketches it would be even possible from the facts learnt by examining

many successive transverse sections, to give the longitudinal course taken by a single nerve-fibre. By pursuing the same mode of enquiry, probably much might be learnt concerning the development and growth of morbid adventitious structures. Those of a soft pulpy character would require to be hardened in chromic acid fluid (pp. 46, 49) before attempts were made to obtain very thin sections, one after the other.

113. Of the use of the Compressorium.—Thin sections may be made thinner by the use of this instrument. Not unfrequently is it required to tear up delicate portions of tissue under the field of the microscope, or to float them out, as it were, from the general substance under examination. In examining minute living animals we often wish to fix them in a position for careful observation. These objects are effected by the compressorium, pl. VII, fig. 7, which consists simply of a mechanical arrangement, by which we are enabled to apply a certain amount of pressure upon the thin glass covering an object, this pressure being increased or diminished at pleasure, and regulated as may be desired. The compressorium consists of a flat piece of brass, with a small ledge on one side, and a large hole in the centre. To one end is attached a lever, bearing a flat brass ring, the rim of which is about a quarter of an inch in width. This ring is free to move, and is capable of being gradually raised from or towards the object, by moving a screw, having a fine thread, which passes through the other extremity of the lever. The compressorium I have just described is one of the simplest forms I have seen, and presents this great advantage:—the ordinary glass slide, with a preparation upon it, may be subjected to pressure without the latter being removed, and placed upon the glass, which, in most compressoriums, is fixed in the hole cut out of the brass plate. Professor Quatrefages has introduced an improvement by which either side of the object can be subjected to examination. The late Mr. Beck improved upon the plans previously introduced, and arranged a compressorium which works wonderfully, even when very thin glass is used. One of these instruments, which was made for me by Messrs. Powell and Lealand, has been of the greatest service in some of my investigations.

114. Of Cutting Thin Sections of Hard Tissues.—Bone and teeth, the hard calcareous plates in the walls of arteries and in cysts, and morbid growths of bony consistence, are, in the first place, cut into thin slices with a fine saw, fig. 5, pl. VII. These are reduced in thickness with a file, or by rubbing on a stone until the sections are transparent, when the scratches may be removed by grinding on a smooth hone with a little water, and subsequent polishing on a dry hone or piece of plate glass, or on a leather on which putty powder has been placed. See also H. to W., §§ 146 to 152. This plan of making thin sections I have, however, long given up. All that was to be learnt from its use

has, I believe, been learnt long ago. I have adopted a plan resting on very different principles. I have found that the hardest bone and tooth structures become, in a measure, softened by being steeped for a long time (from two months to a year or more) in glycerine. I do not know the precise nature of the change that ensues, but the tissue is rendered less brittle and is so altered that it can be cut with a good sharp knife. In this way sections, of course of small extent only, can be removed, and many will be found sufficiently transparent for microscopical examination, even under high powers. A greater degree of softening may be effected by soaking in glycerine to which a few drops of acetic acid have been added (three to five drops to an ounce of glycerine). This will not dissolve away much of the earthy matter. In cases in which it is desired to remove a considerable portion of the calcareous salts, the soaking may be carried out in a fluid consisting of one part of strong hydrochloric acid to five or six of glycerine. Before examination, the specimen ought to be soaked for some time in pure glycerine. Very instructive specimens may be made by partially dissolving away the calcareous matter in acid glycerine. See Chapter VII.

115. Of making Minute Dissections and of Dissecting under Water.—Many very ingenious stands, tables, or stages, have been devised for the purpose of facilitating the operation of dissecting tissues for microscopical examination. The dissecting microscope of Professor Quekett is one of the most useful, and this has been improved upon by Mr. Highley and others. The instrument recently proposed by Mr. James Smith is extremely useful and compact, but the binocular dissecting microscope arranged by Professor Lawson, is the most perfect instrument of the kind yet introduced. For a good form of dissecting microscope, recently arranged by Mr. Swift, see pl. III, fig. 5.

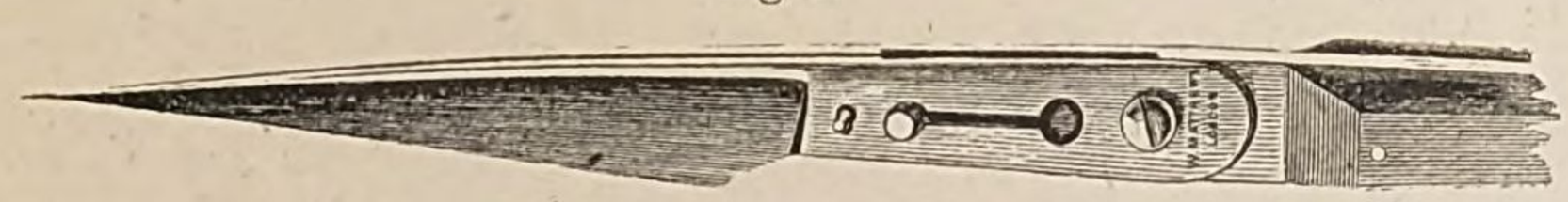
The observer may make a very efficient dissecting microscope by removing the body of the ordinary microscope and replacing it with a bar carrying a simple lens. If the object to be dissected be transparent, the light may be transmitted through it from the mirror beneath the stage of the microscope. If opaque, the light is to be condensed upon it by the aid of an ordinary bull's-eye. Two slanting rests for the arms are easily made of a convenient height with a little thin deal board or mill-board. See also H. to W., §§ 20, 22.

Using the Microscope for Dissection. — In using low magnifying powers, especially for the purpose of dissection, an erect image is obtained if a second object-glass be fitted above the one attached to the body of the microscope. A low magnifying power may be adapted below the stage of the microscope. This arrangement answers admirably for making minute dissections. The microscope must be inclined and the object to be dissected fixed upon cork, in a little trough, or placed upon a piece of plate glass on the table. With the aid of needles and

Fig. 1.

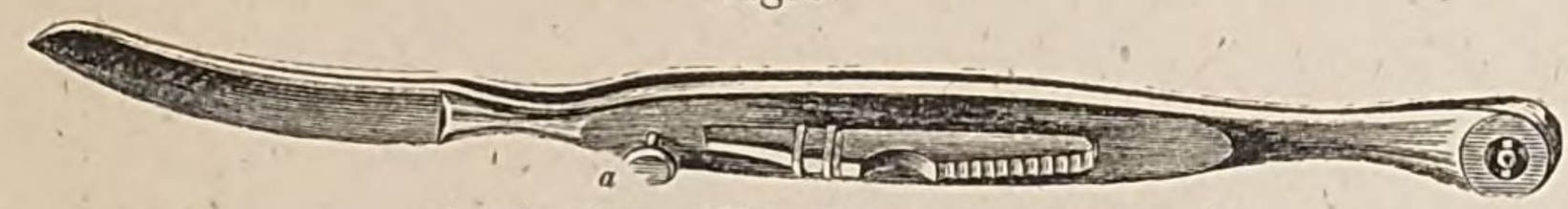
Double-edged scalpel, for cutting thin sections of soft tissues. p. 74.

Fig. 2.



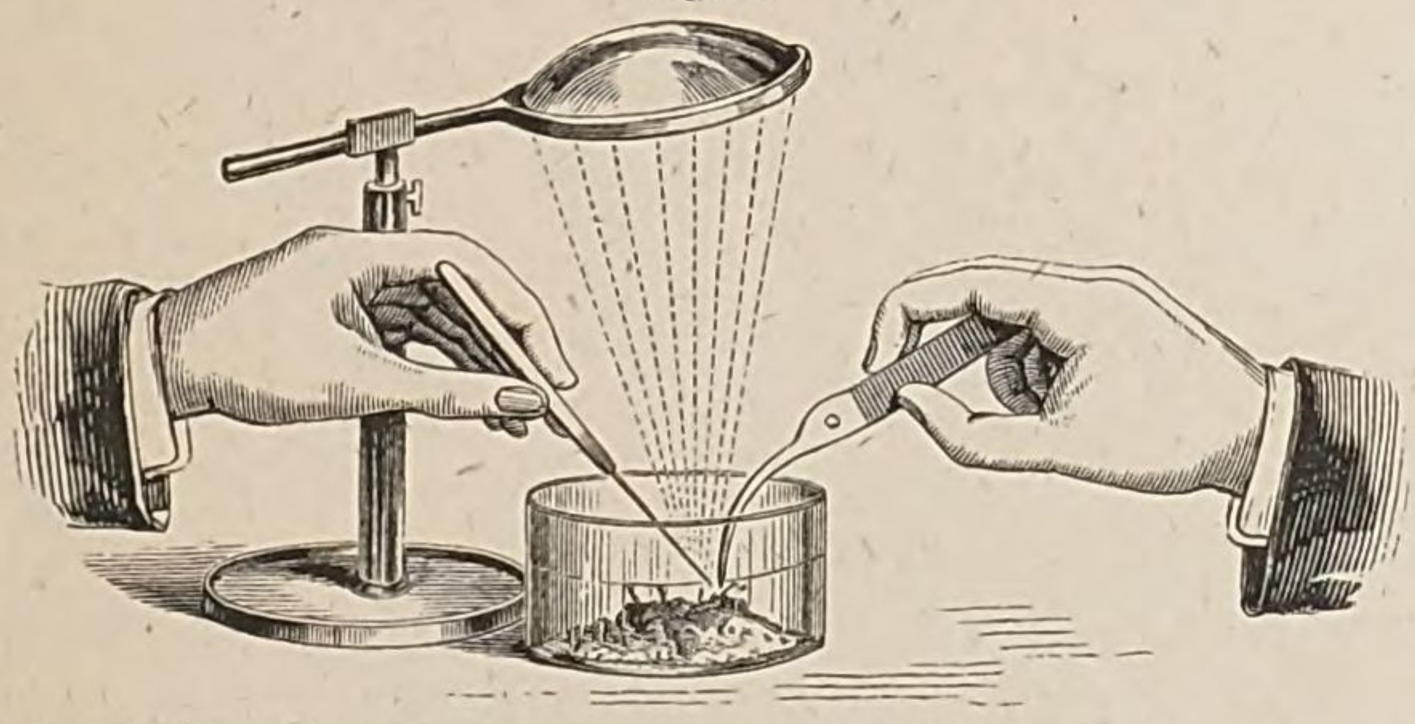
Valentin's knife, for cutting thin sections. p. 74.

Fig. 3.



Another form of Valentin's knife. p. 74.

Fig. 4.



Arrangement for dissecting objects under water The bull's-eye condenser is much larger than that usually employed. p. 76.

Fig. 5.

Fine saw, for cutting thin sections of bone and teeth, which are afterwards to be ground thinner on the hone and polished as described in p. 77.

Fig. 6.

Loaded cork, for pinning objects upon, for the purpose of dissection. p. 80.

Fig. 7.

Compressorium, for pressing out flat thin sections of tissues before mounting. p. 77.

[To face page 73.

forceps, a very careful dissection can be conducted very easily. Mr. Richards has devised a convenient arrangement for fitting the lowest of the two object-glasses in its proper position. A piece of brass plate, of the same size as the ordinary plate-glass slides (three inches by one inch), but somewhat thicker, has in its central part an aperture to which a screw is tapped to suit that of the object-glass. The slide, with the latter screwed into its under surface, is moved into its position and, being properly centred, the instrument is ready for use.

Minute dissections of many tissues are carried on most advantageously under the surface of fluid with the aid of small scissors, needles, or small knives and forceps. If the preparation has been preserved in spirit or other solution, it must be dissected in the same fluid, but in ordinary cases clear water may be used. The microscopist should be provided with a few small dishes, varying in size, and about an inch or more in depth. The large built cells make very good troughs for dissecting in, and circular vessels are made for the purpose, but a common flat pomade pot answers as well as anything, and the cover protects the specimen from injury by dust. In all cases, it should be borne in mind, that as much is gained by condensing a very strong light upon the dissection as by magnifying the object itself.

Watchmaker's Lens.—A very useful instrument to the microscopist is a common watchmaker's loup or lens. With a little practice, the observer will learn to hold it in his eye without any painful effort, and he may then make minute dissections upon a glass slide, or in cases in which it is necessary to fix the object with pins, upon a tablet of cork, or, what is better, wax or gutta-percha. The tablet may be attached to a piece of lead foil, cut to any size desired, and the dissection carried on beneath the surface of water, spirit, or glycerine, as may be desirable, in a small cell. Small tablets, not more than an inch in diameter, will be of great use to the student.

Loaded Corks.—Any object to be dissected under the microscope should be attached to a loaded cork by small pins. We may take a flat piece of cork, rather smaller than the cell, and cut out a piece of sheet lead somewhat larger than the cork. The edges of the lead are then folded over the cork and beaten down slightly with a hammer, and may afterwards be filed smooth with a coarse file, pl. VII, fig. 6.

Of Dissecting a Specimen.—The object being fixed upon the cork and placed in the cell, fluid is poured in until it just covers the surface of the specimen. A strong light is then condensed upon it by means of a large bull's-eye condenser, or by a large globe full of water. With a strong light, magnifying glasses are often not required; and I have always found that delicate dissections could be made with the greatest facility without the aid of a dissecting microscope, if a very strong light was condensed upon the object. Occasional examination of the dissec-

tion with a lens of low power is advantageous; but if the lens be employed during the dissection, there is a great danger of accidentally injuring the specimen, as it is impossible to judge of the distance which the needle point may be beneath the surface of the fluid. Minute branches of nerves or vessels may in this way be followed out, and small pieces of the different tissues into which they can be traced may be removed for microscopical examination with a pair of fine spring scissors. I had scissors of a particular form made, some years since, for very delicate dissections, and I have found them of great use in dissecting thin membranes from delicate structures beneath, pl. VI, fig. 6. With the aid of these scissors, the coats of an artery may be dissected off, and mucous membrane may be readily stripped from subjacent tissues. By a similar plan, the nervous system of the smallest insects can be very readily dissected. The arrangement for carrying on minute dissections is shown in pl. VII, fig. 4.

Tablets upon which Dissections may be Pinned out.—Many preparations require to be arranged in a particular position, before being mounted as permanent objects. *Slabs of wax* are usually employed by anatomists for this purpose, but if transparency is required, the dissections may be attached by threads to thin plates of *mica*.

I have found that the best slabs are made of a mixture of *wax* and *gutta-percha*, in the proportion of one part of the former to from two to four of the latter, according to the degree of hardness required. The ingredients are to be melted in an iron pot, over a clear fire, and well stirred. When quite fluid, the mass may be poured upon a flat slab, and allowed to cool. Thin cakes of about the eighth of an inch in thickness are thus obtained, and they can be easily cut with a knife to fit the cells intended for the preparation. Pins, or small pieces of silver wire, may be inserted into these slabs, and will adhere firmly, although the material is very thin.

116. Of Taking Casts.—Although neither cast-taking nor modelling belongs to the province of the microscopist, many a pathological student will find the advantage of having a knowledge of these operations. A coloured model of a morbid growth can be made very readily, and will show many points that it would be difficult or scarcely possible to give in a drawing. I, therefore, append some excellent directions, given by Mr. Boyd Dawkins, in "Nature," 1872, for taking casts. The plan is simple and well adapted for many objects of natural history and pathological growths. With a very little practice, the student will be able to carry out the operation without difficulty. The material of the mould is artists' modelling wax, which is a composition akin to that which is used by dentists. And as it becomes soft and plastic by the application of heat, though in a cold state it is perfectly rigid, it may be applied to the most delicate object without injury. The method

which has been very carefully described by Mr. Boyd Dawkins is briefly this :—

1. Cover the object to be cast with a thin powder of steatite or French chalk, which prevents the adhesion of the wax.

2. After the wax has become soft, either from immersion in warm water or from exposure to the direct heat of the fire, apply it to the original, being careful to press it into the little cavities. Then carefully cut off the edges of the wax all round, if the under cutting of the object necessitates the mould being in two or more pieces, and let the wax cool with the object in it, until it be sufficiently hard to bear the repetition of the operation on the uncovered portion of the object. The steatite prevents the one piece of the mould sticking to the other. The original ought to be taken out of the mould before the latter becomes perfectly cold and rigid, as in that case it is very difficult to extract.

3. Then pour in plaster of Paris, after having wetted the moulds to prevent bubbles of air lurking in the small interstices, and if the mould be in two pieces, it is generally convenient to fill them with plaster separately before putting them together.

4. Then dry the plaster casts either wholly or partially.

5. Paint the casts in water colours, which must be *fainter* than those of the original, because the next process adds to their intensity. The delicate shades of colour in the original will be marked in the casts by the different quantity of the same colour which is taken up.

6. After drying the cast, steep it in hard paraffin. The ordinary paraffin candles, which can be obtained from any grocer, will serve the purpose.

7. Cool and polish the cast by hand with steatite.

The result of this process is far better than that obtained by any other. The whole operation is very simple, and promises to afford a means of comparison of natural history specimens in different countries, which has long been felt to be a scientific need. It has been already introduced into America and India by Mr. Dawkins, and samples of the casts are to be seen in the British Museum, as well as in that of the Geological Survey, and of Oxford, and of the Queen's College. Casts of typical specimens may be multiplied to any extent, at a small cost of time and money, and are as good as the original for purposes of comparison, and almost as hard as any fossil.

The modelling wax can be purchased from Messrs. Lechertier, Barbe, and Co., artists' colourmen, Regent Street.

Casts of Microscopic Specimens.—By making casts of microscopic specimens, facts may sometimes be ascertained which cannot be demonstrated by examining the specimens themselves. There are inequalities of surface in the case of many delicate tissues that may be rendered

very evident by making a cast in gelatine, in varnish, or in collodion. Casts of the most delicate structures may be obtained with the aid of the latter material. As it takes the most minute markings and striations of the original to which it is applied, the microscopic structure of the surface of the original is faithfully reproduced in the cast. The surface of a moist specimen, of which a cast is desired, must be freed from adhering moisture and left only damp. The transparent cast may afterwards be tinted with a little magenta or other colouring solution, and examined as an ordinary microscopic specimen. By this same process, substances adhering to the surface of natural objects may be removed and the surface thus freed from foreign bodies. The latter, if it be so desired, may be separately examined. Microscopic hair-like appendages of the surface may be attached to the film, and withdrawn with it from the tissue in which they were growing. We may in this way prove whether certain appearances indicative of tissues, such as nerve-fibres, projecting free from a general surface, result from actual structure or are due to some optical effect.

CHAPTER VI.

On Injecting Tissues for Microscopical Examination.—Of Transparent Injecting Fluids.—Prussian Blue Fluid.—Turnbull's Blue.—Carmine Injecting Fluids.—Of the Pressure Required for Injecting.—Apparatus for Injecting by Continuous Pressure.—Of the Operation of Injecting.—On Injecting Morbid Growths.—Of Keeping Injected Specimens for subsequent Examination.

As regards both healthy and morbid structures, many points of great importance can be made out only if injected preparations are examined. From their extreme tenuity and perfect transparency, the capillary vessels are scarcely distinguishable. Indeed, if we examine uninjected preparations only, we may sometimes be led to conclude that a tissue is only slightly vascular, when in fact it is abundantly supplied with vessels. In other cases we may describe as a fibrous matrix or supporting framework, a tissue which is really almost entirely composed of a dense network of capillaries. Capillary vessels when uninjected usually collapse, and in the manipulation necessary for preparing a microscopical specimen, may be pressed and somewhat stretched and torn. Under such circumstances, vessels may be mistaken for nerve-fibres, or for fibrous or connective tissue, and in not a few instances they have been so described.

In connection with the anatomy of morbid growths, still greater confusion has resulted from the vessels of specimens not having been properly injected, before examination under the microscope. It is hardly to be expected that we shall be able to ascertain the nature of the texture, or the history of the various stages through which a particular structure has passed in the course of growth, unless the arrangement of the vessels has been accurately made out, and their precise relation to the most important anatomical elements of the tissue, accurately demonstrated. It is obviously impossible to determine these points, unless the vessels are filled with some coloured material, which renders them more visible than in the natural state. It is at the same time quite clear that they ought not to be filled with any opaque, granular matter, because this would prevent the possibility of the surrounding structures being clearly seen at the same time. For investigations, therefore, of this nature, the materials formerly employed for injecting the capillary vessels, such as vermilion, chromate of lead, and other opaque colouring matters, are unsuitable. Neither can the tissues be dried and mounted in

Canada balsam in the manner in which vascular preparations used to be preserved, because delicate, but most important structures are invariably altered and destroyed by this process, or rendered invisible. In such dried preparations, vessels lying upon very different levels all appear on the same plane, and it is impossible to distinguish whether or not one particular branch of tube or fibre is really continuous with another, or whether the apparent continuity results merely from one lying exactly over or under the other. Many erroneous statements have resulted from observers trusting too implicitly the appearances observed in such specimens.

It is somewhat strange that some minute anatomists of high authority have expressed themselves against this process of injection for the investigation of the anatomy of tissues. It is true that, for the most part, these objections refer to preparations made with opaque materials, the disadvantages of which I have fully explained in my classes since 1854. But so far from the process of injection not being of advantage, I am quite satisfied that more important facts will be learnt from its use than from any other known method of investigating the anatomy of the tissues of animals. The importance of different media for examining tissues is admitted by all, and how, it may be asked, can these be so successfully introduced as by injecting them into channels from which their absorption, by tissue elements, must be almost immediate? By this plan all parts of the tissue will be uniformly acted upon by a solution of one strength. I cannot help fearing that many fanciful objections raised to this mode of investigation have been insisted upon by persons who cannot, or who will not inject, and who are therefore not in a position to form a correct judgment. I am certain that anatomists who do not inject tissues will add little indeed to our knowledge of the minute structure and mode of formation of the tissues and organs of man and the higher animals, either in health or in morbid states. Opposition to the process of injection not only retards true progress, but gives encouragement to the revival of erroneous views which have been abandoned by some of the most distinguished practical anatomists who have preceded us.

117. Properties of the Injecting Fluid.—In order to inject satisfactorily the most minute vessels of a tissue, and at the same time to demonstrate their relation to adjacent structures, we must be provided with an injecting medium which possesses the following properties:—1. The fluid must be of such a consistence that it will run readily through the smallest vessels. 2. It must contain sufficient colouring matter to render the arrangement of the vessels distinct, but must, at the same time, be sufficiently transparent to admit of the examination of the specimen by transmitted light. 3. The colouring matter ought not to be dissolved, because soluble colours often permeate the tissues and

colour all indiscriminately. In this way the vessels may not be readily distinguished from other textures. 4. But though insoluble, the particles of which the colouring matter is composed should be so minute as not to exhibit distinct granules when examined under the highest powers, for granular matter of any kind gives to the specimen a confused appearance. 5. The fluid in which the colouring matter is suspended, must be capable of permeating the walls of the vessels with tolerable facility. 6. It must possess a certain refractive power, and a density greater than that of the fluid which surrounds the tissues in the natural condition. 7. It must be of such composition that it may be used without the previous application of heat. 8. The injecting fluid must not escape too readily from the numerous open vessels necessarily exposed when a thin section of the tissue is removed for examination, and particles which accidentally escape ought not to adhere intimately to the surface of the section, because the specimen would be thereby rendered confused and indistinct, especially if high magnifying powers are required. 9. The fluid employed must not interfere with the preservation of the specimen, and it ought not to undergo any alteration if kept for months. 10. It should be cheap and capable of being readily prepared. 11. The specimens of tissue injected must permit of examination with the highest powers yet made, that is the $\frac{1}{25}$ and $\frac{1}{50}$.

118. Transparent Injecting Fluids.—While searching for a fluid possessing all the advantages above enumerated, a vast number of careful experiments were made. The fluid which I employed in my investigations upon the anatomy of the liver (1854), was found to possess the various qualities required. It is adapted for making minute injections of the capillaries as well as lymphatics and the ducts of glands. This fluid consists of Prussian blue in a state of very minute division, suspended in a solution which acts the part of a preservative fluid. The particles of blue are quite insoluble, so that they will not pass through basement membrane, but at the same time they are so minute that, when examined by a very high power, the precipitate appears uniform and homogeneous. It is not easy to wash this fluid out of the vessels when a section of the injected tissue is prepared, because it becomes incorporated with the serum remaining in the vessels without being precipitated. It runs very freely, and a perfect injection can be made with it in the course of a few minutes. It is well adapted for injecting morbid growths, and possesses many advantages over other injecting fluids of great importance to practitioners who have little time at their disposal for such work. It can be kept for a length of time without being impaired, and can be used at once. Before injecting the tissue no warming is necessary, as in the use of size injections, and the preparation may be examined immediately after the injection has been completed. The fluid is inexpensive, so that small portions of an organ

may be efficiently injected, the loss of a considerable quantity of the injecting material not being a matter of any importance. It tends to harden the coats of the vessels as it passes through their channels, while at the same time it increases the transparency of the specimen. The colour is not affected by acids, and after having been removed by alkalies, may be immediately restored upon the addition of an acid. Capillaries thus injected may be examined by the highest object-glasses.

In using this fluid, it is not even necessary that the pipe should be tied in the vessel, for when this cannot be effected readily, the injection may be driven as the pipe lies loosely in the channel. Although a good deal escapes, much will run in, and the capillaries may often be well injected in this manner. Good injections may be made of small pieces of liver and kidney, although much cut, and in various directions.

Since I first employed transparent injecting fluids, now more than twenty years ago, they have come into general use, and have completely displaced the opaque injections. But although largely employed here and on the continent, it is astonishing how few anatomists seem yet to fully recognise the advantages to be gained by injecting specimens before subjecting them to microscopical investigation. The idea still prevails that the great object of this process is to demonstrate the arrangement of the vessels, but, as I have taught for many years, facts of far greater importance than this may be gained from specimens properly prepared by injection. This matter, however, will be more fully explained in Chapter VII.

It would be a waste of time were I to answer all the ridiculous assertions which have been made by opponents to special methods of investigation which I have found valuable and have recommended, but I will caution the student not to pay too much attention to theoretical objections that are continually being raised. It has been suggested that the injecting fluids recommended by me are excellent when a perfect injection is not desired, while it is a fact that I have specimens thus prepared which are so completely injected that they may be examined with a $\frac{1}{25}$ or $\frac{1}{50}$ of an inch object-glass, and, in at least one instance, the objector has himself seen and examined some of my specimens. One can only feel astonished at such observations. Some authorities seem to consider it necessary to intimate to their pupils, that other observers may be so blind or so stupid that neither what they may have seen nor what they have delineated is to be relied upon, and that their specimens show anything but what they themselves have seen in them. Any intelligent careful student will soon find out that, as regards many so-called anatomical facts, dogmatic assertions have been made for the purpose of destroying real observations which do not happen to accord with the dicta of some infallible authority.

119. Prussian Blue Injecting Fluid.—In former editions of this

work, I have given two receipts for making Prussian blue injecting fluid, one for ordinary injections, the other for the finest injections only. When this fluid was first made Price's glycerine was a very expensive substance, and I thought it well to recommend a cheaper substitute. This is, however, no longer necessary, and I therefore omit altogether the old receipt. The amount of tincture of iron and ferrocyanide of potassium may be doubled if it is desired to inject the small arteries of a very dark blue colour, but I think the observer will find it preferable in most cases to use the fluid of the composition given. Sometimes I add half-an-ounce or more of alcohol, particularly when the capillaries are very tender.

The tincture of perchloride of iron is recommended because it can be readily obtained and is of uniform strength. It is the so-called *muriated tincture of iron*, sold by all chemists and druggists.

The Injecting Fluid.—The following mixture has succeeded admirably in my hands, and I therefore recommend it strongly. It penetrates to the finest vessels. The specimens injected with it retain their colour perfectly, and it can be used in cases in which it is desired that the bioplasm of the injected tissues should be stained with carmine.

Price's glycerine, 2 oz. by measure.
Tincture of perchloride of iron, 10 drops.
Ferrocyanide of potassium, 3 grains.
Strong hydrochloric acid, 3 drops.
Water, 1 oz.

Mix the tincture of iron with one ounce of the glycerine; and the ferrocyanide of potassium, first dissolved in a little water, with the other ounce. These solutions are to be mixed together very gradually in a bottle, and are to be well shaken during admixture. The iron solution must be added *to* the ferrocyanide of potassium. Lastly, the water and hydrochloric acid are to be added. Sometimes I add a little alcohol (one or two drachms) to the above mixture. For the perchloride of iron, five grains of *sulphate of iron*, and for the ferrocyanide of potassium, ten grains of the *ferridcyanide* may be substituted.

This fluid does not deposit any sediment, even if kept for some time, and it appears like a blue solution when examined under high magnifying powers, in consequence of the insoluble particles of Prussian blue being so very minute. This is the injecting fluid I have long used for injecting tissues to be examined under the $\frac{1}{25}$ and $\frac{1}{50}$. It readily penetrates into the finest vessels, and, if one part of a minute capillary is not pervious, it will pass into any minute fissures there may be in the substance that blocks it up. I have specimens of vessels undergoing development in which the injection reaches quite up to the commencement of the portion not yet become tubular.

120. Turnbull's Blue.—My friend, Mr. B. Wills Richardson, of Dublin, has introduced Turnbull's blue and prefers it to the ordinary Prussian blue of my injecting fluid. The other ingredients are the same. Ten grains of pure sulphate of iron are to be dissolved in an ounce of glycerine, or better, in a little distilled water and then mixed with glycerine, and thirty-two grains of ferridcyanide of potassium in another small proportion of water, and the solution mixed with glycerine. These two solutions are then gradually mixed together in a bottle, the iron solution being added to that of the ferridcyanide, and mixture ensured by frequent agitation. This modification may be adopted in cases in which I have recommended the ordinary Prussian blue. The proportions given in the text are unnecessarily large, and I find that the following makes a good fine injecting fluid :—

Ferridcyanide of potassium	10 grains.
Sulphate of iron	5 „
Water	1 ounce.
Glycerine (Price's)	2 ounces.
Alcohol	1 drachm.

The iron, dissolved in a little water and mixed with glycerine, is to be added to the solution of the ferridcyanide, as in the preparation of the other fluid.

Soluble Prussian Blue may be purchased at the drysalters, so that it is not necessary to give the receipt for making it, as the process is troublesome and my book already contains more receipts than the student can possibly use with advantage. Five parts may be dissolved in 100 parts of water, and mixed with 100 parts of Price's glycerine. It must be prepared fresh when wanted. I prefer, however, one of the blue injecting fluids given above.

121. Carmine Injecting Fluid.—In the hands of Mr. A. Smee, Professor Gerlach, and others, carmine has long been employed for making minute injections with the most satisfactory results. The solution is made by adding a few drops of *liquor ammoniæ* to a little carmine, when a beautiful violet coloured *solution* is produced. This may be diluted to the required tint, and injected. It is adapted for injecting very delicate vessels, as those of the brain ; but, if much force be employed, the fluid transudes through the walls of the vessels, and tinges all the neighbouring tissues indiscriminately. The fluid is much improved, and its tendency to transude diminished, by the addition of glycerine and a little alcohol. I had long wanted a transparent injection which could be used for injecting some vessels, while others, in the same preparation, might be injected with Prussian blue. Professor Gerlach has made some beautiful double injections of the portal and hepatic capillaries, by injecting one set of vessels with carmine and the other with Prussian blue. One of these he

kindly sent me by my friend, Dr. Harley, but as Professor Gerlach's preparations were dried and mounted in Canada balsam, there are many important points in the structure which could not be made out. If it is attempted to preserve such a preparation in the moist state, it soon becomes destroyed. The alkali of the carmine injection always destroys the blue colour of the Prussian blue, while if acid be added to the carmine previously, a precipitate unfavourable for injecting the capillaries is produced. After trying a great many different combinations to effect this object, I arrived at the following, which answers the purpose exceedingly well, but it must be made very carefully, or the carmine will be precipitated. *The best carmine only must be used.*

The best carmine	5 grains.
Glycerine, with about eight or ten drops of hydrochloric acid	½ ounce.
Glycerine	1 „
Alcohol	1 drachm.
Water	6 „
Ammonia, a few drops.	

Mix the carmine with a few drops of water, and when well incorporated, add about five drops of *liquor ammoniæ*. To this dark red solution, about half an ounce of the glycerine is to be added, and the whole well shaken in a bottle. Next, very gradually, pour in the acid glycerine, frequently shaking the bottle during admixture. Test the mixture with blue litmus paper, and if not of a very decidedly acid reaction, a few drops more acid may be added to the remainder of the glycerine, and mixed as before. Lastly, mix the alcohol and water very gradually, shaking the bottle thoroughly after adding each successive portion, till the whole is mixed. This fluid, like the Prussian blue, may be kept ready prepared, and injections may be made with it very rapidly.

Dr. Carter's Carmine Injecting Fluid.—For a carmine injecting fluid which will run perfectly freely through the most minute capillaries, and one that will not tint the tissues beyond the vessels themselves, the following formula has been given by Dr. Carter, who has found it answer very satisfactorily :—

Pure carmine	60 grains.
Liq. ammon. fort. (P.L.)	120 „
Glacial acetic acid	86 minims.
Solution of gelatin (1 to 6 water)	2 ounces.
Water	1½ „

The carmine is to be dissolved in the solution of ammonia and filtered, if necessary. With this an ounce and a half of the hot solution

of gelatine is to be thoroughly mixed. The remaining half ounce of gelatine is to be mixed with the acetic acid, and dropped, little by little, into the solution of carmine, the mixture being stirred briskly during the whole time.—("Archives of Medicine," vol. iii., p. 287).

This fluid is admirably adapted for specimens which are to be mounted in Canada balsam, but not for those to be preserved in glycerine. The vessels are well displayed, but all the delicate nerve fibres are invisible.

Generally, my own experience has led me to prefer the blue fluid for injecting, and I use the carmine for the purpose of demonstrating the bioplasm only. See Chapter VII.

122. Other Coloured Injecting Fluids.—Transparent injecting fluids of several different colours are very much to be desired, but although many experiments have been made in the hope of obtaining such, I fear, practically we are restricted to two, the blue and the red. Thiersch, it is true, has succeeded in making others, the composition of one of which, yellow, is given below. I have not, however, met with much success hitherto in the use of these fluids, for if I employ them according to the directions given, I am unable to demonstrate the masses of bioplasm (nuclei), and other points of importance. If made according to the principles followed in the case of the Prussian blue fluid, the results are by no means satisfactory, and as the colour is, in many cases, affected by acids, the subsequent steps of my process are interfered with.

An injecting fluid of a greenish tint may be made, according to the directions given in page 88, for Turnbull's blue, by employing different proportions of the ingredients,—1 grain or less of the sulphate of iron to 10 grains of the ferridcyanide of potassium.

Thiersch ("Das Mikroskop," von Dr. H. Frey) prepares a transparent yellow injecting fluid as follows :—

A.—A solution of bichromate of potash is made, in the proportion of 1 part of the salt to 11 of water.

B.—A solution of nitrate of lead of the same strength.

One part of solution A is placed in a small basin and mixed with 4 parts of a concentrated solution of gelatine. Two parts of solution B are placed in another basin and mixed with 4 parts of jelly.

These are to be slowly and thoroughly mixed together at a temperature of from 75° to 90°, and then heated in a water-bath at a temperature of about 212° for half an hour or more. The mixture is then to be carefully filtered through flannel.

Colouring of the vessels and neighbouring tissues may be produced by staining, as I have stated in page 66. The action of various chemical reagents upon the tissues is ensured by injecting the solutions into the

vessels. In no other way can the elementary parts of the tissues be so thoroughly surrounded with the fluid, or a more uniform action ensured. In Chapter VII, I have described the process which in my hands has been so useful for demonstrating the arrangement of the bioplasm. Recently many observers have adopted the plan of injecting the vessels with weak (from $\frac{1}{4}$ to $\frac{1}{2}$ per cent.) solutions of nitrate of silver, chloride of gold, and other substances (page 69). The student may try solutions of different strengths, according to the degree of staining required. After injection distilled water must be introduced into the vessels, and then $\frac{1}{2}$ per cent. solution of common salt; or, what I prefer, a mixture of one part glycerine to two of water.

123. Of the Pressure required for Successful Injection.—The requisite amount of pressure for forcing the injection into the finest capillaries may be obtained in several different ways. 1. By connecting with a reservoir, placed on a shelf or suspended at a height of two or three feet above the table, a long tube, to one end of which is attached a small piece of India-rubber tube furnished with a stop-cock which fits into the injecting pipe. 2. By placing the injecting fluid in a vessel three or four feet above the table and immersing a syphon tube which may be entirely composed of India-rubber, or partly of glass. 3. By arranging a glass vessel upon the principle of the wash-bottle, p. 107, or the dropping-tube pl. VIII, fig. 2, pressure upon the surface of the liquid being produced, *a*, by the aid of an India-rubber bottle compressed by a weight or spring, or, *b*, by pouring mercury into the tube which reaches nearly to the bottom of the flask. The other tube must of course also dip below the surface of the injecting fluid while to its upper free end a piece of India-rubber tubing provided with a stop-cock at its extremity, must be adapted.

Apparatus for Injection by Continuous Pressure.—This plan, as applied to ordinary injections, was originally proposed by Ludwig, but the same principle was employed by the old anatomists more than 200 years ago in injecting with mercury. A reservoir, from the lower part of which passes a tube having stop-cock and injecting pipes at the end, containing the injection, may be suspended at any height above the specimen, according to the force required; or a series of Wolff's bottles may be used, and into the first water may be conducted, from a reservoir placed at any height above according to the pressure desired. The compressed air acts through a connecting tube upon the injecting fluid contained in the next bottle, which is driven through the issuing pipe previously connected with the duct or vessel to be injected. This apparatus may be modified in many ways. Mercury may be employed instead of water; or a column of mercury may be made to act upon the air on the surface of the injecting fluid. An India-rubber bag may be distended with air, or air may be blown into the vessel containing the injecting

fluid through a tube with a valve. A very simple but thoroughly good arrangement of this kind was designed by Mr. English of St. George's Hospital. The requisite pressure may also be obtained by compressing with a weight a large bag of air from which a pipe furnished with a stop-cock proceeds to the vessel containing the injection. The student who employs this apparatus for continuous injections will find no difficulty in fitting it up for himself.

Other arrangements have also been proposed, but after having tried many different plans, I find that upon the whole, the ordinary injecting syringe is really as successful as more complicated arrangements, while it is cheap, very convenient, and simple, and is most easily kept in perfect order. It need scarcely be said that by no mechanical means can such delicate alterations in pressure be obtained as readily as by the aid of the muscles of the hand and arm. The pressure can be modified instantly, according to the judgment of the operator.

124. Of the Operation of Injecting.—The student will find that the process of injecting will be learnt after a few trials, and although he may quite fail in the first attempts he makes, he is earnestly recommended not to give up, for this mode of investigation is of the greatest advantage, and by it facts are demonstrated of great anatomical importance. Everyone engaged in the investigation of the anatomy of tissues in health and disease, should be able to inject well, and if he employs the fluids recommended, he will find that injections can be made without much sacrifice of time.

The manner in which the operation is performed will now be briefly described. In the first place, the following instruments must be conveniently arranged :—

The syringe thoroughly cleansed with distilled water; with pipes, stop-cock, and corks, all of which must be washed in distilled water just before use, pl. VIII, figs. 4, 5, 6, 7.

One or two scalpels, pl. VII, fig. 1.

Two or three pair of sharp scissors, pl. VI, figs. 4, 5, 6.

Dissecting forceps, pl. VI, fig. 7.

Bull's-nose forceps, pl. VIII, fig. 8.

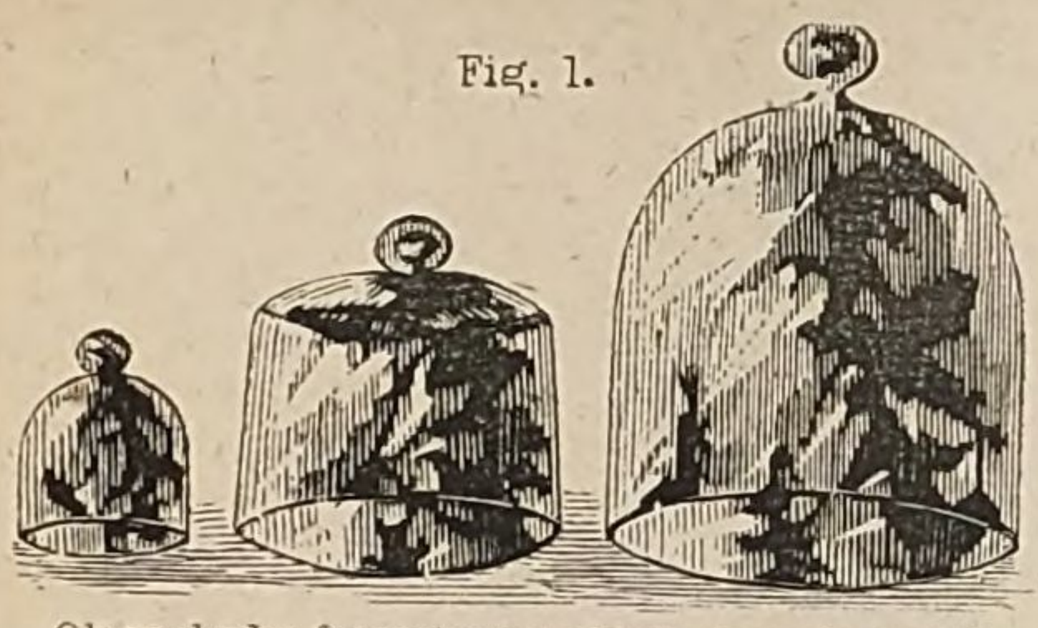
Curved needle, threaded with silk or thread, the thickness of the latter depending upon the size of the vessel to be tied, pl. VI, fig. 9.

Wash-bottle, pl. XVI, fig. 6.

Injecting fluid, a small quantity of which is to be poured out into a small cup or glass beaker which is a little wider than the syringe in its widest part :—

1. An incision is made through the vessel to be injected, with a pair of strong, sharp scissors; the two sides may easily be separated with the aid of a blunt-pointed needle. Into the opening a pipe is inserted and

Fig. 1.

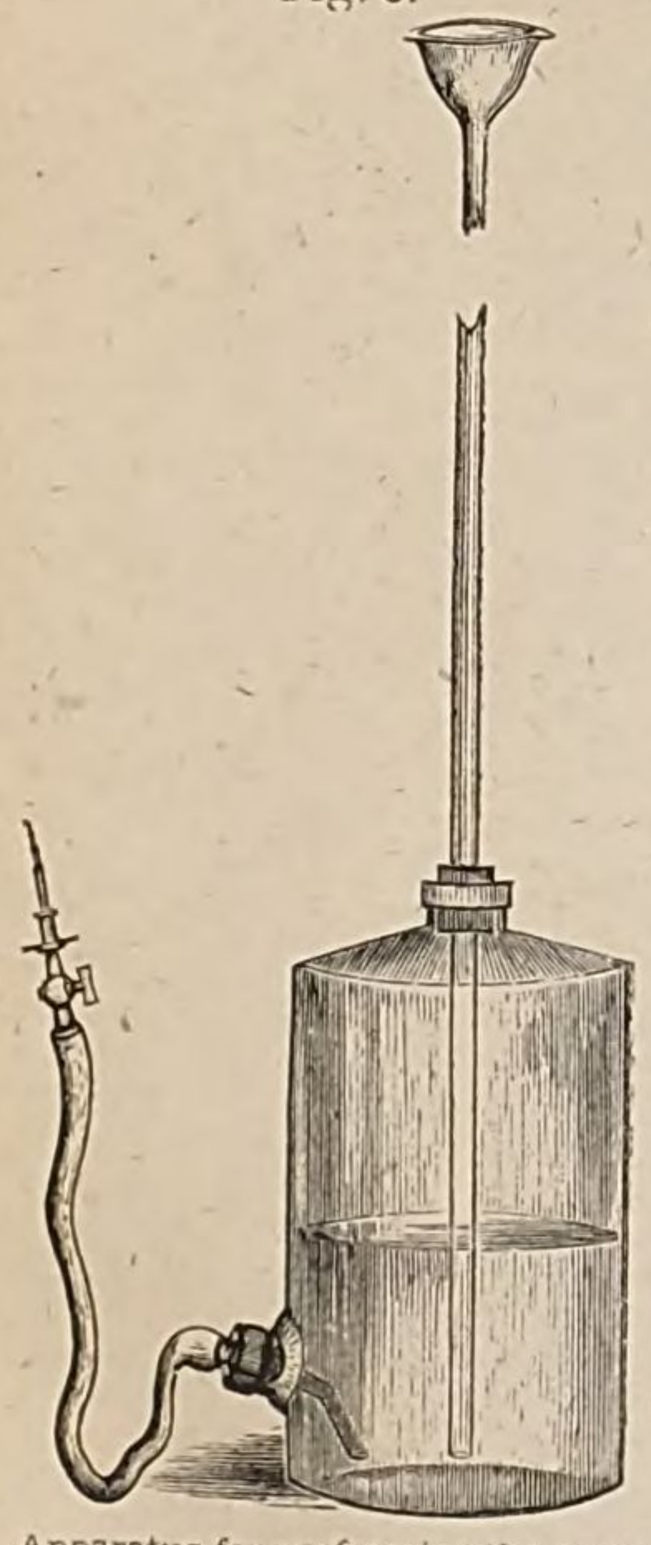


Glass shades for protecting objects from dust while being mounted. p. 56.

Fig. 2.

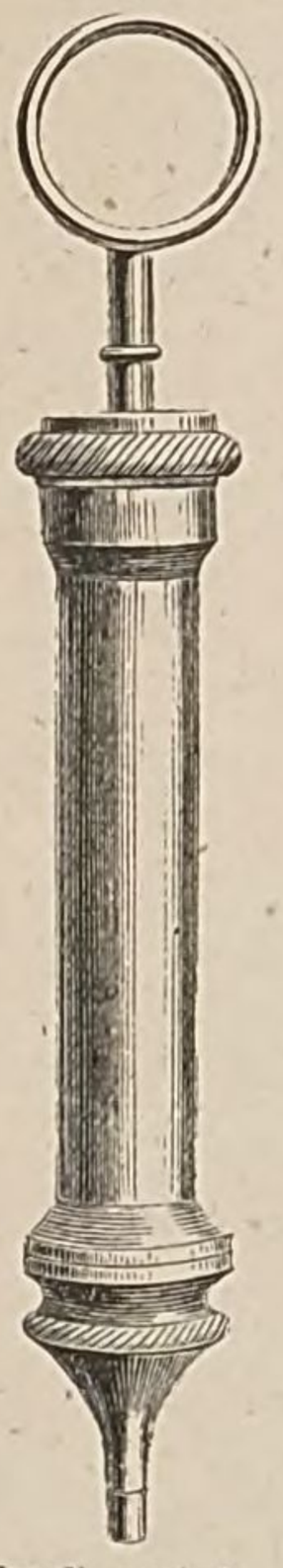
Little bottle for containing glycerine, or glycerine and acetic acid, required for mounting microscopical specimens. p. 107.

Fig. 3.



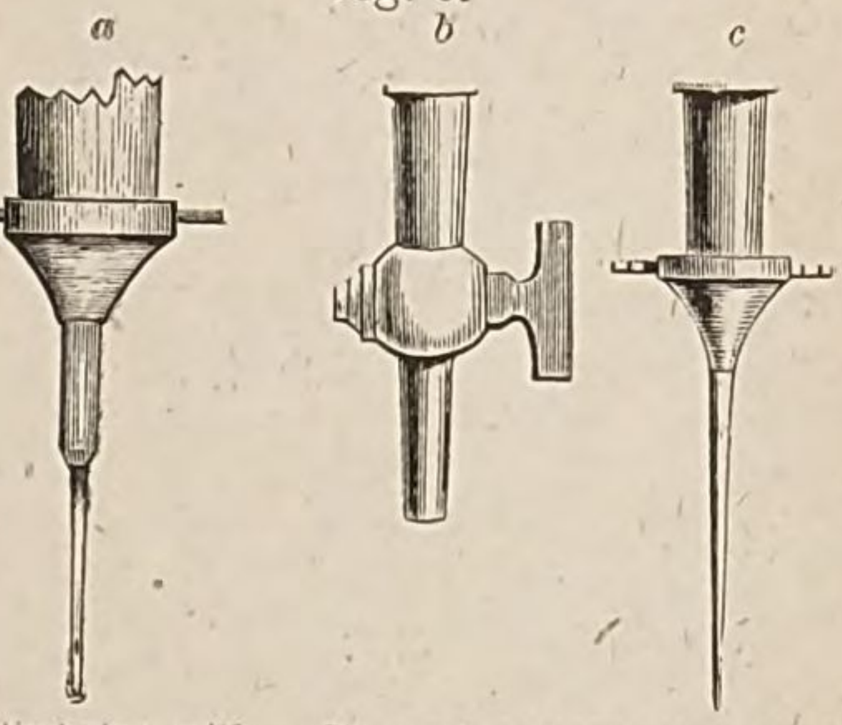
Apparatus for performing the operation of injecting by the pressure of the injecting fluid. p. 91.

Fig. 4.



Small syringe used for injecting. p. 92.

Fig. 5.



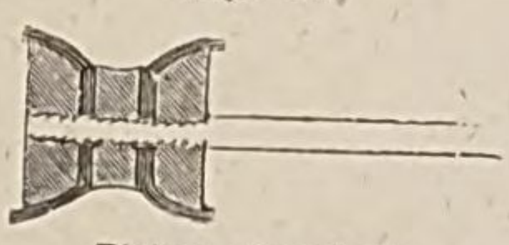
Injecting pipe, with orifice at the side.

Stopcock and injecting pipe, which fit on the syringe. p. 92.

Fig. 6.

Corks for stopping pipes. p. 92.

Fig. 6A.



Piston of syringe.

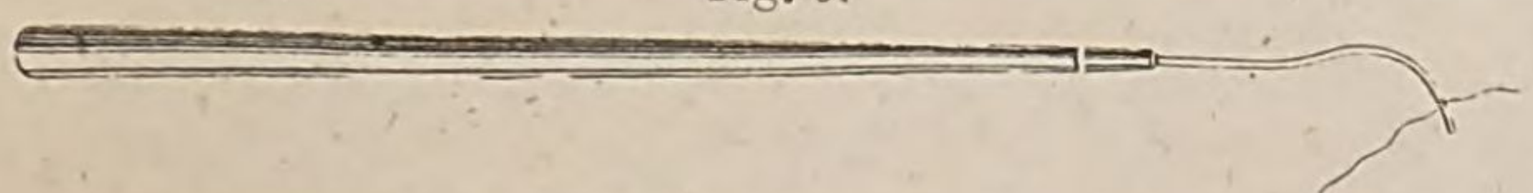
Fig. 7.

Performing the operation of injecting. p 93.

Fig. 8.

Bull's-nose forceps, for stopping vessels from which injection is escaping. p. 92.

Fig. 9.



Needle for passing thread round a vessel which is to be tied upon the pipe.

[To face page 92.

directed towards the point of distribution of the artery. Before the pipe is inserted, however, a little of the injecting fluid is drawn up so as to fill it, or a few drops dropped in it from a pipette, in order to prevent the air contained in the pipe from being forced into the vessels, which would cause the injection to fail.

2. The point of the pipe having been introduced into the artery, the needle with the thread is next carried round the vessel close to its outer surface, and the thread seized with the forceps, and the needle unthreaded and withdrawn; or one end of the thread may be held firmly, while the needle is withdrawn over it in the opposite direction. The thread is now tied over the vessel, so as to include the tip of the pipe only, for if the pipe be tied too far up, there is great danger of its point passing through the delicate coats of the vessel.

3. The nozzle of the syringe is now plunged beneath the surface of the fluid, the piston moved up and down two or three times, so as to force out the air completely, and the syringe thoroughly filled with injecting fluid. It is then connected with the pipe, which is firmly held by the finger and thumb of the left hand, with a screwing movement; a little of the injection having been already forced into the wide part of the pipe so as to prevent the possibility of any air being included.

4. The pipe and syringe being still held with the left hand, the piston is slowly and gently forced down with the right, with a slightly screwing movement, care being taken not to distend the vessel so as to endanger rupture of its coats. The handle of the syringe is to be kept uppermost, and the syringe should never be completely emptied, in case of a little air remaining in it, which would thus be forced into the vessels. The injection will soon be observed running into the smaller vessels in different parts of the structure.

For injecting very minute vessels, glass nozzles may be made very easily by the operator himself out of pieces of glass tube carefully drawn out to the requisite degree of fineness in the spirit lamp. A mark is made with a fine file at a point where the tube is drawn off very fine and carefully broken. The free end is then just heated carefully in the flame to remove the sharp edges, and the pipe is ready for use. The broad end of the pipe is then connected with the injecting apparatus, or with the syringe by a small piece of fine India-rubber tube.

125. Practising Injection.—The student is recommended to practise the process by injecting the organs, and animals, in the order in which they are enumerated, and not to attempt the second until he has succeeded with the first. In all cases the operation is to be conducted patiently, and very slight pressure on the piston is to be exerted.

1. Kidneys of man, sheep, or pig.—*Artery.*
2. Eye of ox.—*Artery.*
3. Rat, mouse, frog.—*Injected from the aorta.* See pl. IX, fig. 1.

4. Portion of intestine.—*Branch of artery.* All divided vessels being tied before commencing to inject. Pl. IX, fig. 2, p. 104.

5. Liver. In one part, a *branch of duct;* in a second, a *branch of artery;* in a third *portal vein;* and in a fourth, *hepatic vein.* The portal and hepatic vein, the artery and portal or hepatic vein, or the duct and portal vein may be injected with injections of different colours in one part.

The branch into which the pipe is to be inserted, must be carefully dissected out in the first instance. When the trunk is small, some difficulty will be experienced in attempting this. The small vessel should be seized in forceps and drawn over the tip of the finger with as little stretching as possible. Next, with a pair of sharp scissors, a slit is to be made quite through the walls of the vessel and slightly extended in a longitudinal direction. If any difficulty is experienced in endeavouring to insert the pipe into the slit, a little water may be projected upon it from the wash-bottle, when the coats of the vessel become slightly raised, and the pipe can be easily introduced into the tube, even if the latter be scarcely wider than the pipe. In injecting very minute vessels, it is better to have the injecting pipe tapering towards a small but blunt point, the hole being at the side, as in the snake's poison fang, instead of at the end.

Where a pipe is required to be inserted into a duct of a gland, which is found with difficulty, even in the natural state of the parts, I have resorted to the following expedient with advantage. Tepid water is gradually injected into the artery or vein which supplies the part. The whole organ swells up considerably, water soon transudes into the follicles of the gland, and passes along the ducts, which, in consequence, become so much distended that they can be easily found; while at the same time, they are completely washed out, and any epithelium or secretion which would interfere with the passage of the injection, removed. An opening can readily be made in the wall, a pipe inserted, and the tube tied over it. When this is done, the water can be removed by wrapping the organ up in cloths and placing the whole under even pressure for twenty-four hours, when nearly all the water will have been absorbed, and the duct and vessels will be in the most favourable state for receiving injection. A pipe may often be inserted into a small lymphatic trunk by pursuing the same course, although it would be quite impossible to introduce it under ordinary circumstances.

126. On Injecting Morbid Growths.—Morbid growths may be injected in the same manner as healthy tissues, but it is almost impossible to obtain very satisfactory results with opaque injections. The walls of the vessels are so thin that they often give way, while the canals themselves are frequently so large and numerous, that when filled with injection other parts of the structure are invisible. In many cases, a very

slight warmth destroys the mass, and reduces it to a mere pulp, so that all injecting fluids which contain size or gelatine are inadmissible.

With the *Prussian blue fluid*, p. 87, I have succeeded in obtaining very good injections of some tumours, and have satisfactory specimens of very soft cancerous growths in which the vessels have been filled without rupture, while the cells are well displayed in the same preparations. From the rapidity with which decomposition takes place, especially in the case of soft tumours, the injection should be commenced as soon as possible after the removal of the tumour. The free passage of the fluid along the vessels is not prevented to the same extent, as in healthy tissues, by the contraction of their coats. Many morbid growths are principally supplied by large veins, which are very readily injected, if only slight force be employed.

Although, as I have already stated, for special enquiries the process of injection by continuous pressure possesses some advantages over the syringe. Injections as perfect and in every respect as good have been made with the aid of the latter simple instrument. One who is always injecting would, of course, keep the last apparatus in good order without difficulty, but the practitioner who only requires to perform such an operation now and then, will, I think, find the small syringe the most useful instrument. The injecting fluids I have recommended and the simple syringe can always be kept ready for use. The injection of a piece of an organ or morbid growth can be completed in the manner I have described, and anything cleaned and put away within half an hour. The injected specimen is put into glycerine for subsequent examination.

127. Other Objects of Injecting Vessels.—The process of injection is employed for other purposes besides demonstrating the arrangement of the vessels of a tissue. The value of various chemical reagents, in hardening textures, and in rendering certain structures transparent, or increasing the opacity of others, has already been alluded to (§§ 91, 92, 93). It is obviously desirable that tissues to be examined in any of these fluids should be thoroughly saturated with them in every part, and no portion should long remain out of contact with fluid of the same composition as that in which it is to be examined or preserved. In the ordinary manner in which tissues are prepared, especially if they be very thick, it is a long time before the fluid in which they are immersed penetrates into the interior, and when it does so, in consequence of having been filtered through a considerable thickness of structure, its action will be weakened. Indeed, in many instances, decomposition will have commenced before the preservative solution can have thoroughly permeated every part of the structure. All these difficulties and inconveniences may be avoided if the vessels of the part be injected with some of the same solution as that in which the preparation is to be preserved, or with a somewhat weaker solution. By such a plan, it is

clear that every portion of the tissue will be bathed with the fluid, which permeates the delicate walls of the capillaries, just as they are permeated by fluid which transudes through them during life, and bathes all the adjacent tissues. Latterly, I have injected various tissues with different preservative fluids and chemical reagents, and have obtained most satisfactory results. The Prussian blue fluid serves the double purpose, not only of rendering the arrangement of the vessels distinct, but of making the tissues transparent, and at the same time it acts the part of a preservative solution. It penetrates so readily that even the interstices in tissues may be injected with it, and it will pass into and occupy the channels between epithelial and other cells.

By pursuing the methods above recommended, many fallacies will be guarded against. Uninjected and contracted capillaries have often been mistaken for a form of fibrous tissue, and the loop-like arrangement of vessels in some textures has produced an appearance which has been mistaken for epithelial cells. Indeed, in the uninjected state, especially when a few blood corpuscles remain in the vessels, the resemblance to an epithelial layer is very striking. Such an appearance is seen both in the lungs and in the kidney. If, however, the vessels are injected with the Prussian blue fluid, or with a colourless solution, the blood corpuscles are washed out, the vascular loops are distended, and the walls of the capillaries are seen in profile, as sharp, well-defined, clear, and unbroken outlines. I have injected fluids which have the property of rendering cells, where they exist, exceedingly distinct, and in no single instance have I ever been able to demonstrate, in either of the above situations in the adult, an epithelial layer such, for instance, as that figured by Kölliker, in his drawing of the air cells of the lung, or in the drawing of the capillaries of the Malpighian body by Dr. Isaacs ("On the Anatomy and Physiology of the Kidney." Transactions of the New York Academy of Medicine, 1857). It has been urged, by those who have described the latter epithelium, that in the progress of injection it is forced off from the vessels, but even if this occurred to some extent, we should certainly find a few cells still adherent, here and there; and it is easy to show that epithelial cells, where they exist, are not so easily removed by injecting the vessels as has been supposed. The delicate epithelium in the minute gall ducts, is not unfrequently found still adhering to the basement membrane, not only after the ducts have been thoroughly washed out with water forced through them in one direction, but after they have been injected with fluid caused to traverse them in an opposite one.

128. Of Keeping Injected Specimens.—Portions of tissue injected with the glycerine fluids I have recommended, may be kept in glycerine with five drops of acetic acid to the ounce, for any length of time, or they may be subjected to examination immediately after the operation of

injecting has been completed. By maceration in acid of glycerine, however, the tissues are rendered a little more transparent and slightly softened — no small advantages for investigating many healthy and diseased structures. The pieces of tissue to be kept for subsequent examination should be placed in little corked tubes, and the strong glycerine added a few drops at a time, on several successive days, for the reasons given in p. 101. The general process of preparation in this Chapter will be found practical, and of special advantage to medical practitioners, because they may be carried out quickly and the specimens set aside without any risk of deterioration, until the observer may have leisure for their careful study. See pl. IX, figs. 3 and 4, for examples of specimens injected as described. The most minute capillaries are fully injected.

After injecting transparent injecting fluids of which gelatine forms the basis, the tissues or organs when cold are to be soaked in methylated spirit, containing one per cent. or less of hydrochloric acid. Sections may then be made and preserved in glycerine, damar, or balsam. The student who adopts this process will often find that the coloured gelatine shrinks considerably, occupying the centre of the vessel, a clear space being left between it and the vascular wall. Such changes have led to many curious speculations regarding structural arrangements, and the strangest conjectures have been offered concerning the correct interpretation of the appearances. The coloured gelatine injected into vessels sometimes penetrates into interstices in tissues and fills the spaces between cells. After it has become cold, and has been placed in alcohol, the gelatine shrinks, the edge presents a defined outline, and the appearance is exactly that which would result if the gelatine had been injected into very narrow capillary tubes with membranous walls, although, in truth, it may be positively proved, in other ways, that no tubes whatever existed in the situation in question.

CHAPTER VII.

The Author's method of preparing Healthy and Morbid Tissues for Examination, under the highest Magnifying Powers.—Principles upon which the Process is based.—Of the Solutions required.—The Plan followed for Tissues generally.—Of injecting the Tissue and Staining the Bioplasm.—Of Examining the Tissues when properly prepared.—New Method of preparing Specimens of Bone and Teeth, and other hard Tissues.—Advantages to be derived from the Process.

In 1863–4 I published some memoirs on minute structure ("On the structure and formation of the so-called apolar, unipolar and bipolar nerve cells of the frog;" "New observations upon the minute anatomy of the papillæ of the frog's tongue," Phil. Trans., 1863–4), in which the great value of higher magnifying powers than had ever been used before in the investigation of the growth and structure of tissues of vertebrate animals was demonstrated. My observations attracted little attention, for they were not in accordance with the "tendency of thought" of the time. By them, however, the real usefulness of powers, magnifying one thousand diameters and more, for the investigation of the anatomical structure of man and the higher animals was distinctly proved, and few workers would now dispute the practical advantage of high powers such as were first made for, and used by me. They have since come into general use in Germany, from which country numerous object-glasses, magnifying from 700 to 1,500 diameters have been imported into England, and it is to be hoped that in the course of a few years anatomical observers may be permitted to make use of high magnifying powers without discredit.

Much misconception prevails concerning the advantages to be derived from the examination of the tissues of man and the higher animals under the powerful objective referred to. Many, and among them a few distinguished anatomists, continue to express themselves averse from the use of these high powers for tissue examination. They say, and with truth, that by increasing the apparent size of an object you do not thereby necessarily discover anything new concerning its real structure and arrangement; but by such a remark the author convicts himself, not only of want of knowledge concerning what has actually been achieved, but exhibits a want of appreciation of the advantages of certain methods of examination by the aid of which new facts have been

revealed, and with which he might have been thoroughly acquainted some years ago.

For a time, and at a period of mental progress, when dogmatic unreasoning assertion constantly repeated wins unreasoning converts in numbers, the conjectural dicta of a clique must necessarily prevail over all individual statements. And though certain anatomical facts may be demonstrated distinctly enough, it unfortunately happens that few Englishmen have sufficient self-confidence to minutely describe what they have seen under a microscope, until they have been assured that what they seem to have seen is not opposed to what dominant scientific authority has affirmed is to be seen and demonstrated, or if not actually to be demonstrated, is nevertheless in accordance with the particular undemonstrated facts which only, according to authority, ought to be accepted and taught; so that for a mere worker who differs from authority to win many supporters in these days in England or elsewhere may be set down as impossible even in thought. I conceive few real workers are foolish enough to waste their time in trying to "convince" anyone.

Still it must be admitted that anyone who has succeeded in making out new facts does enjoy the liberty of being permitted to record them, and need not be deterred from explaining his views, or from giving a detailed account of the methods of demonstration he has found to be of advantage. After having prepared his readers for the contemptuous remarks they will hear concerning his work and his methods, and cautioned them not to be led away from the consideration of facts by the cries or threats of scientific cliques, an individual worker may put on record the results of his experience in the hope of assisting others to become workers, independent of the tyranny of sects.

129. Principles upon which the Process is based.—It is necessary to consider in the first place what circumstances interfere with the perfect demonstration of structure under the highest powers of the microscope, and how the operation of these may be prevented.

1. Of many tissues, sections sufficiently thin for high powers cannot be obtained by the processes usually adopted. In order to make the specimen thin enough, *pressure* must be employed, and in many instances very strong pressure is required. Now, tissues immersed in water are destroyed completely, even by moderate pressure under glass. Experience has proved that unless the tissues are immersed in, and are thoroughly impregnated with, a viscid medium, they cannot be made thin enough by pressure. But a medium to be of real use should be readily miscible not only with water in all proportions, but with such chemical re-agents as may be required to act upon one or more constituents of the tissues for the purposes of demonstration.

2. As many structures are exceedingly delicate, and undergo change

very soon after death, it is necessary that the medium in which they are examined should have the property of preventing softening and disintegration, and should also act the part of a preservative fluid.

3. In order that tissues may be uniformly permeated by a fluid within a very short time after the death of the animal, it is necessary that the fluid should come quickly in contact with every part of the texture. This may be effected in two ways :—

a. By soaking *very thin* pieces of the tissue in the fluid, or
b. By injecting the fluid into the vessels of the animal.

4. As different structures require fluids of different refractive power for their demonstration, the medium employed must be such that its refractive power can be increased or diminished, unless, for the medium fulfilling the former condition, another can be readily substituted which fulfils the latter requirements.

5. In investigations upon the changes which structure undergoes in the organism, we want to distinguish between that part of the texture which is the oldest, and that which has been recently produced—between matter in which active changes are going on, and matter which is in a passive state. The process of carmine staining described in pp. 65, 66, and 104, enables us to demonstrate not only the direction in which growth is taking place, but the precise point where new matter is being added to that which is already converted into structure.

6. It is necessary in many investigations, that the vessels should be positively distinguished from the other constituents of the tissue, and it is important that the process by which this is effected should not interfere with the demonstration of nerves or any other tissues in the immediate vicinity of the vessels.

7. It is necessary that the medium employed for demonstration should have the property of preserving the specimens, so that observers may be able to exhibit to others the actual specimens upon which their observations have been made.*

Glycerine and *syrup*, with certain colouring matters, fulfil the requirements mentioned in the foregoing paragraphs. It is necessary in many cases to employ the strongest glycerine. In this country we have had the advantage of the beautiful preparation called Price's glycerine, which is made of specific gravity 1240. Strong syrup may be made by dissolving, with the aid of heat, lump sugar in distilled water.

It has been said that glycerine and strong syrup are not adapted for preserving soft tissues, because the tissues shrink and soft cells collapse

* The process of investigation described in this section was first published in the third edition of "How to Work with the Microscope," August, 1864, but the author had adopted it for four years previously.

in consequence of osmose of their fluid contents. But I have many hundred specimens preserved in the strongest glycerine I could procure, and I should obtain advantages if glycerine could be made of still greater density. There would be no difficulty in impregnating even very soft tissues with it. Tissues possess a highly elastic property, and although they shrink when immersed in a medium of great density, they gradually regain their original volume if *left in it for some time.* In practice, the specimen is first immersed in *weak* glycerine or syrup, and the density of the fluid is gradually increased. In this way, in the course of two or three days, the softest and most delicate tissues may be made to swell out almost to their original volume. They become more transparent, but no chemical alteration is produced, and the addition of water will at any time cause the specimen to assume its ordinary characters.

The hardest textures, like bone and teeth, may be thoroughly impregnated and preserved in strong glycerine; and the softest, like cerebral tissue, delicate nervous textures like the retina, or the nerve textures of the internal ear, may be permeated by the strongest glycerine, and, when fully saturated with it, dissection may be carried to a degree of minuteness which I have found impossible when using any other medium. Nor is the use of glycerine and syrup confined to the tissues of man and the higher animals. I have preparations from creatures of every class. The smallest animalcules, tissues of entozoa, polyps, star fishes, mollusks, insects, crustacea, various vegetable tissues, microscopic fungi, and algæ of the most minute and delicate character, as well as the most delicate parts of higher vegetable tissues may all be preserved in these viscid media; so also may the slowest and most rapidly growing, the hardest and softest morbid growths, as well as embryonic structures at every period of development, even when in the softest state. I am, indeed, not acquainted with any animal or vegetable tissue, which cannot with the greatest advantage be mounted thus. All that is required is, that the strength of the fluid should be increased very gradually until the whole tissue is thoroughly penetrated by the strongest that can be obtained. Glycerine has long been in use among microscopists. It is universally applicable, and glycerine or syrup may be made the basis of all solutions employed by the microscopical observer with the greatest advantage. Many points, which have hitherto escaped observation, are to be demonstrated by the use of these solutions, and it is certain that very much will yet be discovered by the aid of these media.

130. The Solutions required.—From these general remarks, I pass on to describe, more in detail, the particular methods I have adopted during the last fourteen years, for minute investigations upon structures magnified by the highest powers yet employed. It will be necessary,

in the first place, to give the composition of the different solutions which I find useful for general purposes.

1. *Weak glycerine* of about the specific gravity 1050.

2. *The strongest Price's glycerine* that can be obtained.

3. *Syrup* made by dissolving, by the application of a gentle heat in a water-bath, 3 lbs. of sugar in a pint of distilled water. A weaker solution can be prepared, as required, by mixing equal parts of syrup and water. The tendency to crystallization may be prevented to some extent by the addition of a little acetic acid, but this objection to the use of strong syrup cannot be completely overcome, while if dilute syrup be used, its refractive power is not sufficient, and fungi are very prone to grow in it and destroy the specimens.

4. *Prussian blue injecting fluid and carmine fluid.*—These fluids should be kept ready prepared. They will keep well for months. The composition of the blue injecting fluid is given on p. 87, and that of the carmine fluid on p. 65, and the glycerine solutions required after the staining process is complete, on p. 66. The blue injecting fluid is required for enabling us to distinguish the vessels, even the most minute capillaries, in the specimen intended for examination, and the carmine fluid is necessary for the clear demonstration of the bioplasm or living matter of the several tissues—a point of the utmost importance in studying the structure, formation and growth of tissues, and the changes which occur in them in disease.

The composition of these fluids has been arrived at after numerous experiments, and the correct proportion of glycerine ascertained by repeated observations. The method of procedure recommended must be adopted as a whole. It consists, as will be noticed, of several distinct steps, each of which has reference to the rest, and must be conducted with the same care.

It having been established as a principle that, for minute investigation, tissues must be immersed and thoroughly saturated with viscid media miscible in all proportions with water, it almost follows that re-agents applied to such tissues should be dissolved in media of the same physical properties. For a long time past I have been in the habit of employing solution of potash, acetic acid, and other re-agents, dissolved in glycerine instead of in water. In some cases I have found the addition of very strong solutions of certain re-agents necessary. For example, the greatest advantage sometimes results from the application to a tissue of very strong acetic acid. If the acid be added to glycerine in quantity, the solution will no longer be viscid, so that another plan must be resorted to. I thicken the strongest acetic acid with sugar, a gentle heat being applied to dissolve the sugar. Thus a very strong acetic acid solution of the consistence of syrup can be most readily prepared. Strong solutions of potash, soda and other

re-agents are to be made in the same way. Thus a complete chemical examination may be conducted upon tissues, solutions, or deposits preserved in *viscid* media. The reactions are most conclusive, but of course take a much longer time for completion than when carried out in the ordinary manner. Ten or twelve hours must be allowed to elapse before the change is complete, the process being expedited if the slide be placed in a warm place (about 100°). A preparation after having been made in acetic acid syrup may be transferred to glycerine and acetic acid without undergoing much alteration, but it must be soaked in the glycerine for some days before it is mounted.

5. *Chromic Acid Fluid.*—A fluid most valuable to the microscopist is a solution of chromic acid in glycerine, and another solution of bichromate of potash in the same fluid. A few drops of a strong solution of chromic acid may be added, so as to give to the glycerine a pale straw colour. The bichromate of potash solution is prepared by adding from twelve to twenty drops of a strong saturated solution of bichromate of potash to an ounce of the strong glycerine. By this plan, the hardening effects of these re-agents upon the finest nerve tissues are improved, while the granular appearance which is caused by aqueous solutions of these substances is much diminished. Sometimes advantage seems to result from mixing a little of the chromic acid with the acetic acid solution of glycerine.

If desired, sugar may be substituted for glycerine in all the fluids employed, including the carmine and injecting fluids; but, as already stated, a great inconvenience results from the tendency to growth of fungi, especially in warm weather. Camphor, creosote, carbolic acid, naphtha, prevent this to some extent; but it is a disadvantage from which strong glycerine is perfectly free. Sometimes, too, crystallisation occurs, and destroys the specimen. In using first a syrupy fluid, and then glycerine, to the same specimen, it must be remembered that the two fluids mix but slowly, so that plenty of time must be allowed for the thorough penetration of the medium used last.

I keep various tests, such as alcohol, ether, the various acids, and alkalies, and other tests in the form of viscid solutions made with glycerine or sugar. The reaction of the iodine tests (Chapter X, § 198) for amyloid matter, starch and cellulose, is much more distinct when employed in this manner. The plan is, to allow the texture to be tested, to be thoroughly saturated with the strong glycerine solutions, and then to add water. In the course of a few hours the reaction takes place very strongly.

131. The plan practically followed for preparing Tissues of Vertebrata.—The general plan I follow, is the same for all tissues of all vertebrate animals and morbid growths; but I will describe the several steps of the process as they were conducted in the demonstration of the minute

structure of ganglion cells, and of the structure of the papillæ of the frog's tongue (Phil. Trans., 1863–4). The description given also applies to the mode of preparing specimens of muscular fibre to demonstrate the mode of distribution of the finest branches of nerve fibre, and specimens of the minute structure of the brain, spinal cord, and ganglia of man and the higher animals.

Perhaps it will be most useful to describe the mode of proceeding when a frog is to be prepared for minute injection. My researches upon the tissues of the frog have been principally conducted upon the little green tree frog (Hyla arborea), for experience has proved to me that the tissues of this little animal are so much more favourable for investigation than those of the common frog, that it is well worth while to obtain specimens, even at the cost of *2s.* or *2s. 6d.* each. The student may, however, obtain very beautiful specimens from the common frog.

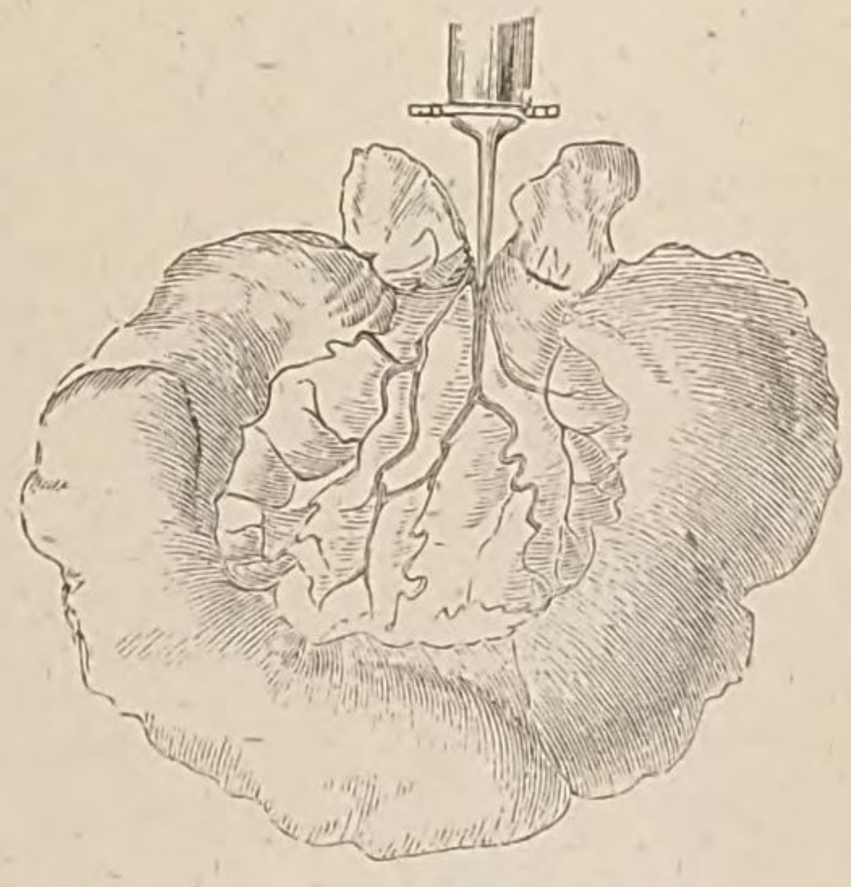
Of injecting the frog.—The animal is carefully folded up in a large cloth, which is then suddenly dashed upon the floor. In this way sudden death is ensured without the tissues being bruised in the least degree. An opening is immediately made in the sternum, the heart exposed, and its apex cut off. Through the opening a fine injecting pipe, after being filled with a little injection is passed and directed into the artery, the vessel being carefully tied to the point of the pipe, as directed in page 93 (2). See also plate IX, fig. 1. The operation of injection ought to be completed in from twenty minutes to half an hour, and sometimes may be effected in less time than this. The injection being pale, cannot be very distinctly seen by the unaided eye, but if the operation has been conducted successfully, the tissues will be found swollen and the areolar tissue about the neck of the animal will be fully distended with the glycerine fluid that has filtered through the vascular walls.

Of staining the bioplasm.—The injection being complete, the abdominal cavity of the frog is opened, and the viscera carefully washed with a little glycerine diluted with about one-fourth part of water. For such purposes an ounce or a two-ounce bottle fitted up like the wash-bottle (pl. XVI, fig. 6), will be found very useful. The legs of the frog may be removed, the mouth slit open upon one side, and the pharynx well washed with glycerine. If it is desired to prepare one organ only, this may be removed and operated upon separately; but the entire trunk, with all the viscera, may be subjected to the action of the carmine fluid. If the brain and spinal cord are special objects of enquiry, the cranium and spinal canal must be opened so as to expose the organs completely, before the staining process is commenced. Enough of the carmine solution is then placed in a little porcelain basin or gallipot, just sufficient to cover the entire trunk and viscera. The specimen is then moved about in the carmine fluid, so that every part that is exposed

Fig. 1.

Small hyla or green tree frog, with fine injecting pipe inserted into aorta, ready for being injected. p. 104.

Fig. 2.

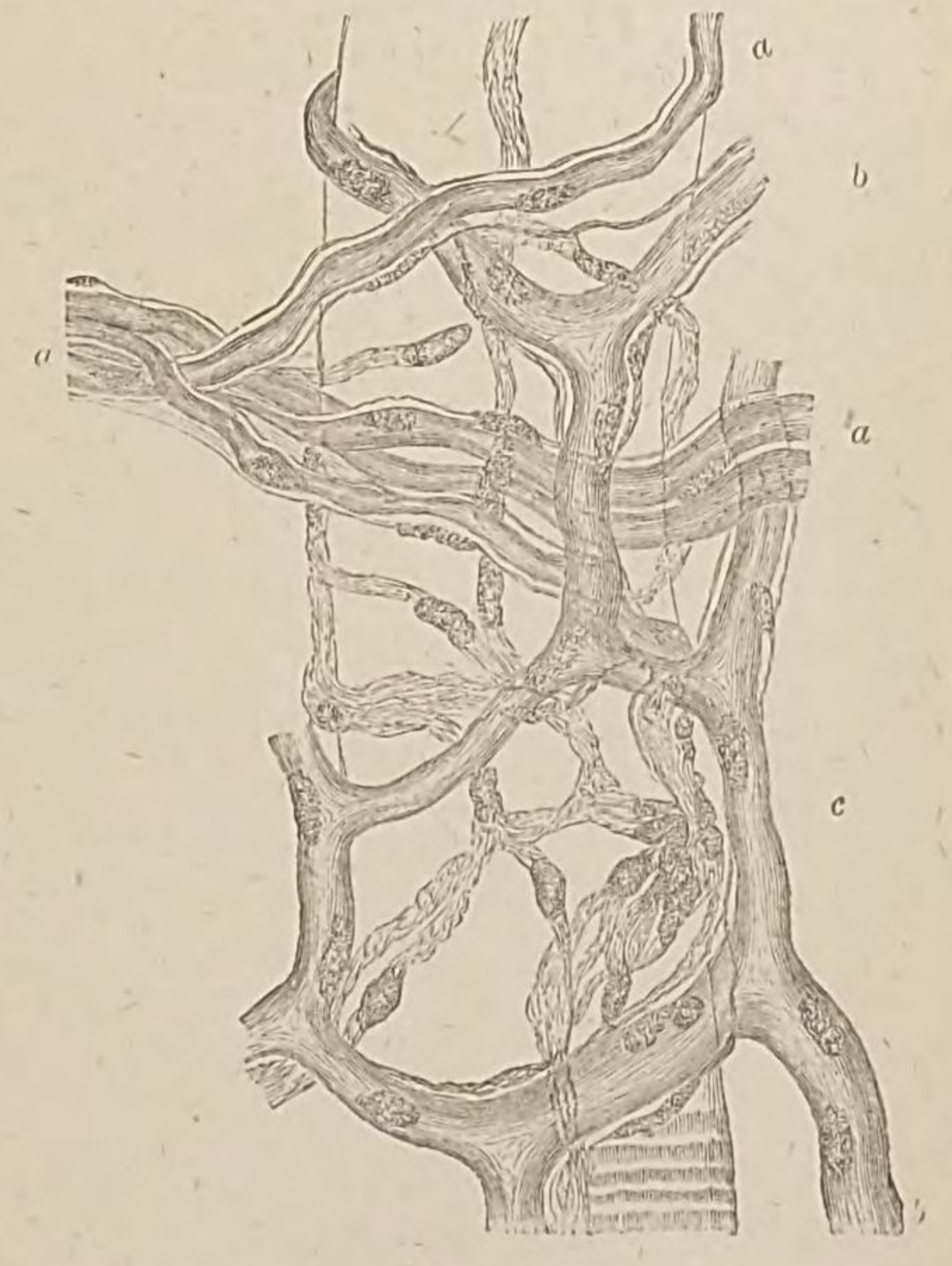


Injecting pipe inserted into one of the arteries of mesentery, so that the vessels of one part of the intestine may be injected. p. 94.

Fig. 3.

Portion of a muscular fibre from the diaphragm of a mouse, showing nerve fibres and capillary, vessels. × 700. p. 111.

Fig. 4.



Capillaries and nerves of a single muscular fibre of the white mouse. The capillaries are well injected. *a*, Dark bordered nerve fibres *b*, Capillary. To the left of *b* and *c* are nerves to capillaries. × 700. p. 111.

[To face page 104

may be thoroughly wetted by it; sometimes slight pressure with the finger is required. It is left in the carmine fluid for a period varying from four to six or eight hours, being occasionally pressed and moved about during this time, *so as to ensure the carmine fluid coming into contact with every part.*

The blue colour of the vessels of the lungs, viscera, &c., will have almost entirely disappeared, and all the tissues will appear uniformly red. The staining is now complete. The carmine fluid is poured off and thrown away, and the preparation washed quickly with the glycerine solution (2 parts of glycerine, 1 part of water). The specimen is now placed in another little basin, and some strong glycerine poured over it; it is then left for two or three hours, and a little more strong glycerine added. When, from six to twelve hours have elapsed since the specimen was removed from the carmine solution, it is ready for the last preliminary operation. The glycerine used for washing it is poured off, and sufficient strong Price's glycerine added just to cover it. To this, three or four drops of strong acetic acid are added, and well mixed with the glycerine. In this acid fluid the preparation may be left for several days, but it should be removed about from time to time, so as to insure every part coming into contact with the acid glycerine. A small piece of some vascular part may be cut off, placed in a drop of glycerine, and subjected to microscopical examination. If the injected vessels are of a bright blue colour, and the bioplasm (nuclei) of the tissues of a bright red, the specimen is ready for minute examination; but if the blue colour is not distinct, three or four more drops of acetid acid must be added to the glycerine, and the preparation soaked for a few days longer.

If the bioplasm is of a very dark red colour, and appears smooth and homogeneous, more especially if the tissue intervening between the bioplasts is coloured red, the specimen has been soaked too long in the carmine fluid; but in this case, although parts upon the surface may be useless for further investigation, the tissues below may have received the proper amount of colour.

Of injecting with carmine and Prussian blue fluids.—Another plan which I have adopted, and which, although more difficult in practice, if carried out with due care, possesses some great advantages, is the following:—The vessels are in the first instance thoroughly injected with the carmine fluid. The preparation is allowed to remain for four-and-twenty hours, in order that the carmine may be absorbed. A little glycerine is then to be injected, and lastly the Prussian blue injecting fluid introduced until the capillary vessels are completely filled with it. The carmine fluid must be injected very slowly, and but slight pressure employed, or the vessels will certainly be ruptured, for the ammonia renders them very soft. When the Prussian blue fluid has been successfully introduced

into the vessels, the textures required for investigation may be removed, washed in glycerine, and after soaking for a day or two in acetic acid glycerine, will be ready for microscopic investigation. Beautiful and most perfect specimens of solid internal organs, like the brain and spinal cord, may be obtained by this process; and it is the most perfect of the many plans I have adopted, although many practical difficulties will be met with in carrying it out. It will probably fail in the hands of the student unless he has the patience to repeat the attempt many times; when, however, success is obtained, the observer will be well rewarded for the trouble he has taken, and for the many failures he may have experienced. He will be provided with tissues the investigation of which would employ him for years.

132. Of examining the tissues when properly prepared.—The tissues or organs to be subjected to special investigation having been removed, and transferred to fresh glycerine, may be kept in little corked glass tubes, pl. XVI, fig. 7, and properly labelled. Generally, the tissue will contain sufficient acetic acid, but if this is not the case, one drop more may be added. The examination should not be commenced until the specimen has been soaked for some days. It may be preserved for a length of time without other change than slight softening of the tissues by which the dissection for microscopic examination is much facilitated.

Suppose, now, the nerves with the small vessels and areolar tissue at the posterior and lower part of the abdominal cavity, have been placed in one tube, and the prepared tongue of the Hyla in another. The former specimen may be taken out of the glycerine, and spread out upon a glass slide. If it be examined with an inch power, numerous microscopic ganglia will be seen. Several of these will be found close to small arteries. Those which are most free from pigment cells are selected, and removed carefully by the aid of a sharp knife, fine scissors, forceps, and a needle point. This operation may be effected while the slide is placed upon the stage of the microscope. The *transmitted light* enables the observer to see the minute pieces very distinctly, but if necessary, a watchmaker's lens may be used, or the dissecting microscope referred to in page 78 employed. The pieces selected are transferred to a few drops of the strongest glycerine placed in a watch-glass or small basin, or in one of the little china colour moulds, and left to soak for several hours, being carefully protected from dust by one of the glass shades, pl. VIII, fig. 1, p. 92.

The microscopical examination of the specimen may now be carried out. One of the small pieces is removed from the glycerine and placed upon a glass slide, in a drop of fresh glycerine, and covered with thin glass. The glass slide may be gently warmed over the lamp, and the thin glass pressed down upon the preparation by slight taps with a

needle point, or small piece of hard wood or ivory terminating in a blunt point. The specimen may now be examined with a quarter, and afterwards with the twelfth of an inch object-glass. A good deal of granular matter and débris may possibly obscure the delicate points in the structure. The slide is again gently warmed, and, with the aid of a needle, the thin glass is made to slide over the surface of the specimen, without the position of the latter being altered, and then removed and cleaned. The specimen is next washed by the addition of drop after drop of strong glycerine containing five drops of acetic acid to the ounce. The slide can be slightly inclined while it is warmed gently over the lamp, in such a manner that the drops of glycerine slowly pass over the specimen and wash away the débris from its surface. The most convenient instrument for dropping the glycerine on the specimen is a little bottle, of two ounces capacity, with a syphon tube drawn to a point, and a straight tube, with an expanded upper part, over which is tied a piece of *stout* sheet vulcanized India-rubber. Upon compressing the air, by pressing down the India-rubber, the glycerine is forced drop by drop through the syphon tube and allowed to fall from a height of three to six inches upon the specimen.* See pl. VIII, fig. 2, p. 92.

When several drops of pure glycerine have been allowed to flow over the specimen, the thin glass cover, after having been cleaned, is re-applied and pressed upon the specimen very gradually, but more firmly than before. If the preparation looks pretty clear when examined with the twelfth, the glass cover may be cemented down with Bell's cement, or with damar solution, and the specimen left for many days in a quiet place. It may then be re-examined, the process of washing with glycerine repeated, and further pressure applied until it is rendered as thin as is desired. When this point has been reached, more glycerine with acetic acid is to be added, and a plate of mica or the *thinnest glass cover* applied, when it may be examined with the twenty-fifth. The process of flattening and thinning may be pushed still further if desirable, —and if this be carried out *very slowly* by gentle taps or careful pressure with the finger and thumb *from day to day*, the elements of the tissues may be gradually separated without being torn or destroyed. If there be much connective tissue, which interferes with a clear view of the finest nerve or muscular fibres, it may be necessary to immerse the specimen for some days in the acetic acid syrup, or soak it in the digestive fluid (§ 90, p. 42), and then transfer it to fresh glycerine. The success of this process depends upon the care and patience with which it is carried out. The most perfect results are obtained in cases where the washing, pressure and warming have been very slowly conducted.

* These little bottles, as well as any other instruments or apparatus required, can be obtained of Mr. Matthews, Carey Street, Lincoln's Inn Fields; or Mr. Swift, University Street, Tottenham Court Road.

It is most interesting to notice the minute points of structure which have gradually become clearer and clearer by the successive application of a gentle heat, subjecting the specimen to a little firmer pressure and by soaking it in a little fresh glycerine placed in a watch-glass.

Specimens of tissue prepared in this way can be transferred from slide to slide, and no matter how thin they may be, after having been allowed to soak in fresh glycerine they may always be laid out again perfectly flat upon another slide, by the aid of needles. I prefer, however, to mount these specimens upon a circle of thin glass about $\frac{3}{4}$ of an inch in diameter, instead of upon a glass slide. The covering glass is of the thinnest kind, and less than $\frac{1}{2}$ an inch in diameter. When this has been properly fixed by cement the circle is placed in a wooden slide in the centre of which a hole has been drilled of the proper dimensions to receive it. The glass circle is fixed in its place and to the wooden slide by a ring of gummed paper. The action of viscid fluids in the investigation of the delicate details of minute structure is most valuable, and I feel sure that by the method of procedure here given, the principle being retained, while the details are modified in various particulars in special cases, as experience may show to be desirable, many new and important anatomical facts will be discovered. Until this process has been carried out by other observers, exactly as I have adopted it, I cannot expect that many of my own observations will be confirmed, and I do not feel surprised that many authorities in Germany, and therefore elsewhere, doubt whether I have seen the things which I have seen and have demonstrated to others.

133. New Method of preparing Specimens of Bone and Teeth and other Hard Tissues.—By the methods generally employed for demonstrating the structure of bone, teeth, and other hard tissues, described in § 114, p. 77, we are enabled to form a notion of the dead and dry tissue only. The soft material is dried up before the section is made.

And yet this very soft material, which is not represented in the drawings published in many standard works, is that which makes the only difference between the dried bone or tooth in our cabinets and that which still remains an integral part of the living body. So far from this soft matter being unimportant, it is the most important of all the structures of the hard texture. It is by this alone that all osseous and dental tissues are formed and nourished, and from the arrangement of this soft matter not having been recognized, the most erroneous ideas have prevailed, and still prevail, upon the formation and nutrition of the osseous and dental tissues.

Even now it is generally believed that the dentinal tubes are real tubular passages for conveying *fluids* to all parts of the dentine, and are thus subservient to its "nutrition," and yet it is more than eight years since Mr. Tomes proved most conclusively that these so-called

"tubes" were occupied in the recent state by a moist but tolerably firm material (Phil. Trans., Feb., 1856). I have verified Mr. Tomes' description, and am quite certain that the so-called dentinal tubes are not channels for the mere flowing up and down of nutrient fluid.*

Suppose a tooth is to be prepared for minute microscopical investigation we may proceed as follows. The same plan is applicable to bone and shell.

1. As soon as possible after extraction, the tooth may be broken by a hammer into fragments, so as to expose clean surfaces of the tissues. Pieces of dentine with portions of pulp still adhering to them may then be selected and immersed in the carmine fluid (p. 65), and placed in a vessel lightly covered with paper, so as to exclude the dust. The whole may be left in a warm room for from twenty-four to forty-eight hours.

2. The carmine solution may then be poured off, and a little plain dilute glycerine added, as described in the case of soft tissues (p. 105).

3. After the fragments of teeth have remained in this fluid for five or six hours, the excess, now coloured with the carmine, may be poured off, and replaced by a little strong glycerine and acetic acid.

4. After having remained in this fluid for three or four days, it will be found that the portions of soft pulp have regained the volume they occupied when fresh. They have swollen out again even in the strongest glycerine.

5. I have found that in many cases, when it is desired to study the arrangement of the nerves, it is necessary to harden the pulp by immersion in a solution, made by adding to an ounce of the mixture of glycerine and acetic acid, two or three drops of a strong solution of chromic acid. The fragments may remain in this solution for three or four days, and then be transferred to the acetic acid solution, in which they may be preserved for years with all the soft parts perfect.

6. The specimens are now ready for examination. Thin sections are *cut* with a knife from the fractured surfaces of the dentine, including a portion of the soft pulp. The knife should be strong, but sharp. In practice I have found the double-edged scalpels made for me by Messrs. Weiss and Son, of the Strand, answer exceedingly well for this purpose, nor will the edge of the knife be destroyed so soon as would be supposed (pl. VII, fig. 1).

7. The minute fragments of sections thus obtained are placed upon a slide and immersed in a drop of pure strong glycerine, in which they may be allowed to soak for an hour or more, and then examined by a low power (an inch). The best pieces are then to be selected by the

* On the structure of recent bone and teeth, see my lectures on "The structure and growth of the tissues." Delivered at the Royal College of Physicians, 1860.

aid of a fine needle, and removed to a drop of glycerine containing two drops of acetic acid to the ounce, and placed upon a clean slide. Lastly, the thin glass cover is carefully applied, and the specimen may be examined with higher powers.

8. If it is desired to retain the specimen, the excess of glycerine fluid is absorbed by small pieces of blotting paper, and the glass cover cemented to the slide by carefully painting a narrow ring of Bell's or damar cement round it. When this first thin layer is dry, the brush may be carried round a second time, and, after the lapse of a few days, more may be applied. Mounted in this way the specimen will retain its characters for years.

Hard tissues, like bone, dentine, and enamel, become softened by prolonged maceration in glycerine, and if a few drops of acetic acid are added, the softening process may be carried to a greater extent, and yet without the calcareous matter being entirely dissolved out. If desired, of course the calcareous matter may be in part or entirely removed by increasing the strength of the acid fluid in which the preparation is immersed. But far short of this, the hard, brittle texture is so altered that thin sections may be *cut* without any difficulty. Specimens prepared in this way may be examined by the highest magnifying powers yet made,—by which statement I mean, of course, to imply that more may be learnt by the use of such high powers (1,000 to 3,000 linear) than by employing ordinary object-glasses.

134. Advantages of the Process.—Of the very many new methods of preparation introduced during the last few years and warmly advocated, only those which I have found most useful have been recommended in this work. Had more been given, I fear I should have confused rather than assisted the student. I have been so disappointed with the practical results of some plans, although most strongly recommended by their inventors, that no good purpose would have been served by their introduction. Indeed, I am satisfied that many of these new methods rest upon unsound principles altogether, and by drawing conclusions from what is very imperfectly demonstrated in the specimens prepared, observers have been and are being led away from the truth, and encouraged to accept views, some of which have been actually proved to be erroneous by preceding observers.

As regards the value of certain methods recently recommended for investigating the distribution of the finest nerve fibres, I can speak confidently. If these processes be adopted, it will be found that the very part of the nerve fibre which is required to be followed is actually destroyed, and thus the observer has been led to conclude that the nerve *ended* very near to the dark-bordered fibre, the truth being, as proved by altogether different methods of enquiry, that the nerve cannot be seen to end at all, and can be traced a very long distance beyond the point

where it is believed to terminate, as a bundle of extremely fine fibres, which are arranged in networks.

Such points as those, clearly delineated in figs. 1 and 2, pl. X, and in pl. XI, would not have been visible in specimens, had any other process of investigation than the one recommended been followed. The delicate nerve fibres ramifying in every part of the connective tissue would not have been seen had the specimen not been prepared according to the principles laid down in p. 99. I have never been able to demonstrate the fine nerve fibres distributed to the capillary vessels, except by the method of investigation I have recommended. In my specimens they can be traced in every part of their course from the nerve trunks to their ultimate distribution, quite close to, and sometimes in actual contact with the vascular walls; and it is possible to distinguish the nuclei or masses of bioplasm connected with these nerve fibres from those belonging to the capillaries themselves. This highly important fact is not seen in specimens prepared by the gold staining process; and although by this latter plan many more fibres come into view than are to be seen in my preparations, it will be admitted that no one has yet proved that all the fibres seen are nerve fibres, while by my plan, the true nature of every one of the fine fibres can be proved beyond a doubt because they can be followed uninterruptedly into an unquestionable nerve trunk. Hence, I can place implicit reliance upon my demonstration as far as it has been carried, while I have reason to doubt the accuracy of the conclusion of those who seem to have carried their researches beyond the point which I have reached in my search for the ultimate ramifications of nerve fibres.

As far as I am aware, such specimens as those which I have represented in pls. X and XI, could only have been prepared in the manner recommended. The delicate branches of the nerve fibres so well seen in pl. XI, would not have been visible had the specimen been mounted in damar or balsam, and the excessively delicate connective tissue could not have been manipulated so as to show the ganglion cell and its fibres unless it had been long soaked in very strong glycerine. In the latter medium such delicate preparations are easily moved about and placed in any position that may be desired. The thin membrane can be made to lie perfectly flat, it can be readily turned over, and may be subjected to considerable pressure if it is not sufficiently thin for examination under high powers. From such a preparation pieces may without difficulty be snipped off with fine scissors, or removed with a sharp knife. Any accidental folds or creases may be easily removed with the aid of needles or pins. I may incidentally mention that I find long pins, and especially steel pins, of great use in manipulating delicate microscopical specimens. They are easily held between the finger and thumb, and I can work with them even better than with needles mounted in handles. Pins are

less likely to slip, and one has perfect command over them. Such specimens I arrange with the aid of a watchmaker's lens, and a couple of pins, in a proper position to be covered with the thin glass, which after having been breathed upon, is allowed to fall gently upon the surface of the drop of glycerine. The other specimens represented in pls. IX and X, and which have been prepared in the same manner, are sufficiently described in the explanations underneath them.

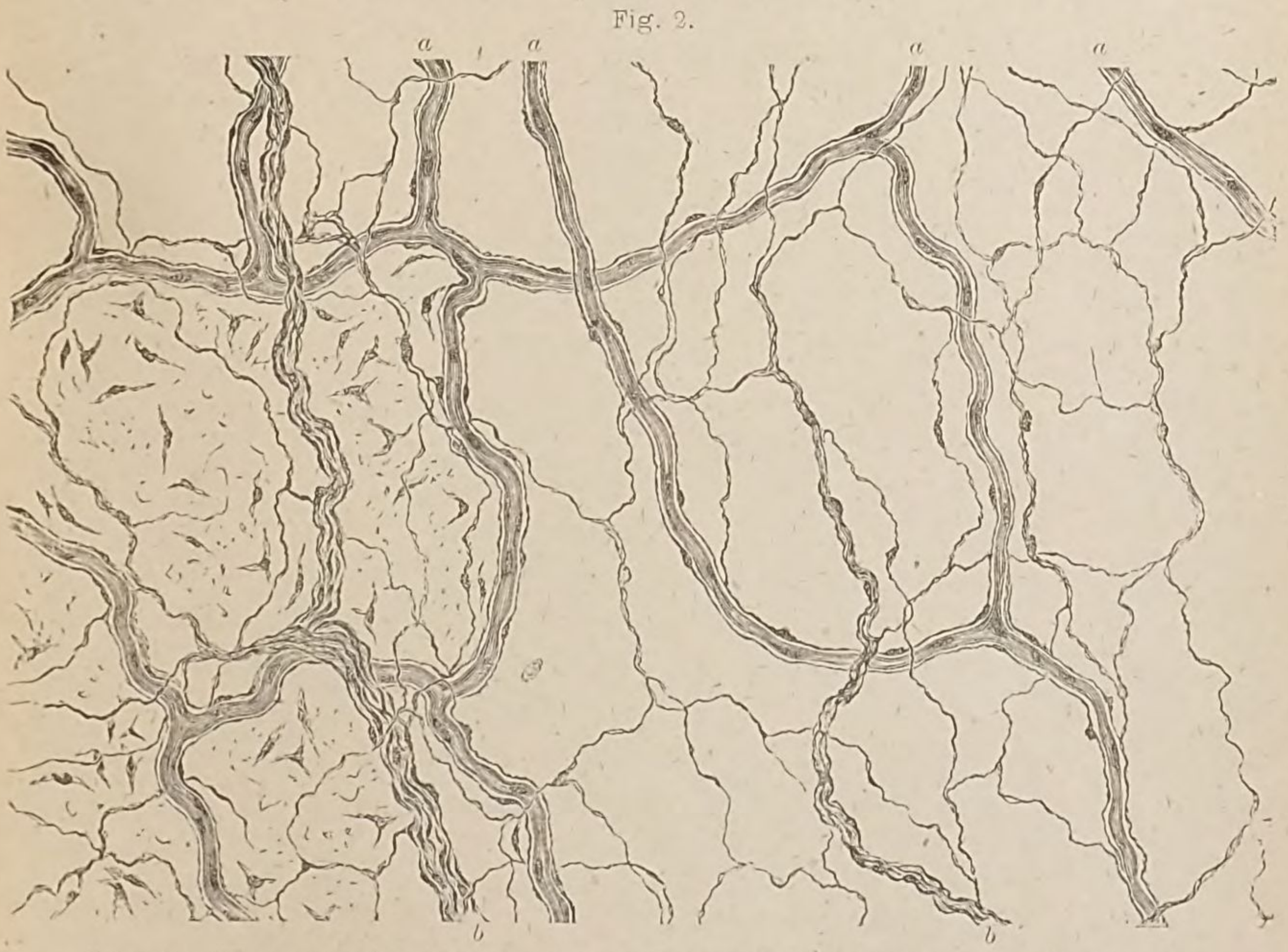
Those who condemn the plan I have recommended, without being practically acquainted with it, and those who seek to excite prejudice against it by denying or ignoring the conclusions I have arrived at, and the truthfulness of my drawings without having seen the specimens, are taking a course which cannot be fair, but which at any rate I shall not object to. As my specimens are preserved and can be examined by any one who cares to do so, it is scarcely necessary to say more. The actual specimens from which figs. in pls. IX, X, and XI were copied, have been mounted for upwards of ten years, and some are still in a good state of preservation. It is, of course, vain to hope that many observers can come here to minutely examine specimens. Various circumstances render this impossible. Time, however, will determine which of several conflicting observations is correct. Fanciful anatomical statements are gradually neglected, and pass quietly out of notice while observations which are real and worth retaining, will be revived and re-examined. They may be modified and corrected, but at last the exact truth concerning many profoundly interesting questions at present very obscure will be found out, admitted, and at length generally received.

By the process of investigation described in this chapter, and which has been followed out by me for about fifteen years, the observer is enabled to learn much that is new concerning the minute structure and arrangement of fully formed tissues, but he may gain besides valuable information upon the growth of many tissues and organs. General facts of the greatest importance with reference to the phenomena of development are also to be demonstrated by preparing the tissues according to the principles laid down. No one who has conducted the anatomical analysis of typical organisms at different periods of development, with the aid of this system of investigation will fail to have acquired facts of the highest value for synthetical reconstruction and arrangement. Any-one who has studied in the mannner recommended for a sufficient length of time may have presented to his intellectual vision that wonderful series of phenomena which succeed one another, but without interruption, from the moment of the formation of the germ to the moment of the death of the fully formed organism. The ever-changing picture which may thus be realized by the understanding, and contemplated by the intellect of the patient observer, it would be impossible even for consummate skill adequately to describe in words or transfer to canvas. The details can be seen

Fig. 1.

Development of nerve fibres, muscular fibres, and connective tissue, very young newt. Prepared according to the author's method for examination under very high powers × 700. *a*, nerve fibres. *b*, muscular fibres. *c*, very young muscular fibres. *d*, capillary vessel. See chap. vii. pp. 98 and 111.

Fig. 2.

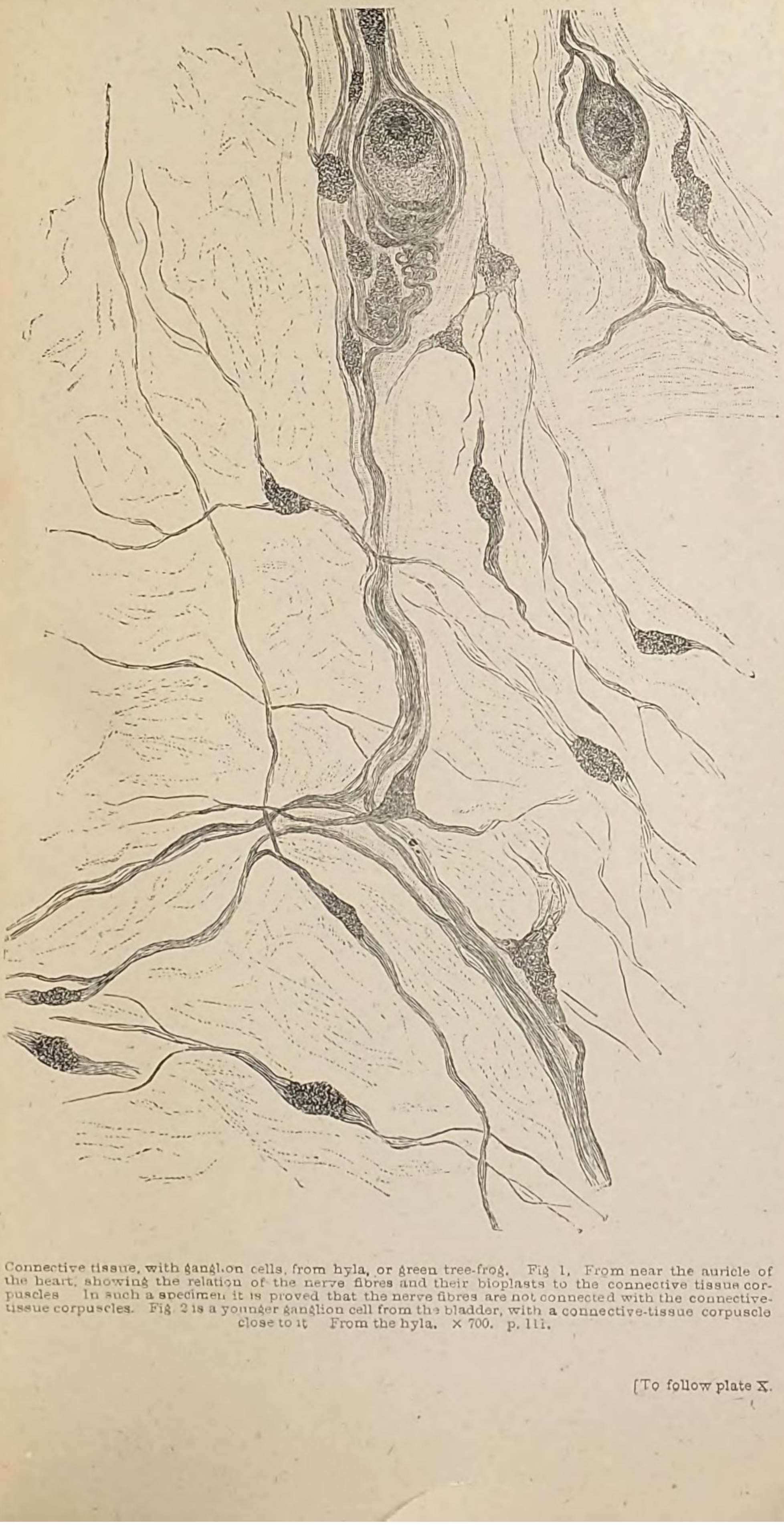


Connective tissue covering mylo-hyoid muscle of the hyla, or green tree-frog. *a*, capillary vessels, with their nerve fibres. *b*, bundles of fine dark bordered nerve fibres, from which fine nerve fibres may be traced to the capillaries and to their distribution in the connective tissue, where they form networks of exceedingly fine compound fibres. The engraving represents the specimen as magnified only 110 diameters, but the original drawing was taken from it when magnified by a much higher power. × 110. p. 111. 1861.

[To face page 112

Fig. 1.

Fig. 2.



Connective tissue, with ganglion cells, from hyla, or green tree-frog. Fig 1, From near the auricle of the heart, showing the relation of the nerve fibres and their bioplasts to the connective tissue corpuscles In such a specimen it is proved that the nerve fibres are not connected with the connective-tissue corpuscles. Fig 2 is a younger ganglion cell from the bladder, with a connective-tissue corpuscle close to it From the hyla. × 700. p. 111.

[To follow plate X.

and contemplated only by those who devote themselves thoroughly to this highly interesting department of knowledge, which I fear is never likely to be generally appreciated, in consequence of the time, skill and patience required for its successful prosecution, and the little credit and reputation to be gained by those who follow it compared with what is to be earned more easily in other departments. The certainty of not reaping any pecuniary reward necessarily deters many from entering upon such labours, and sadly reduces the number of those who, with every desire to study and with love for the work, find it possible to engage in it for a sufficient length of time to excel.

CHAPTER VIII.

Of examining Living Matter in Health and Disease, and Living Beings under the Microscope.—Of compensating for Evaporation.—Arrangements for testing the Influence of Gases, for passing Currents of Electricity through Living Objects, and for maintaining a Uniform Temperature.—Of Anæsthetics.—Of feeding Living Matter.—Of studying the Movements of Bioplasm.—I. Primary or Vital Movements.—Cyclosis.—Centripetal Movements.—Movements in all Directions.—II. Secondary Movements.—Ciliary and Muscular Action.—III. Inorganic Movements.—Vital Movements of Amœba of White Blood-Corpuscle, Mucus, and Pus-Corpuscle.—Movements of the Bioplasm of the Ovum.—Phenomena of the Circulation, and of the Action of the Heart and Arteries during Life.—Circulation in the web of the Frog's Foot.—Lungs.—Heart's Action.—Branchiæ.—Movement of the Chyle.—Phenomena of Growth and Multiplication.—Growth of Roots and Leaves of Vegetable Tissues.—Of the Abnormal Increase and Multiplication of Bioplasm.—Inflammation, a, *Non-vascular Phenomena;* b, *Vascular Changes.—Aeroscopy.—Aeroscopes.—Nature of the Bodies collected from the Air.—Of detecting Solid Particles in the Breath.—Of detecting Ammonia in the Breath.*

He will be the best practitioner who is always trying his utmost to form an accurate conception of the changes which are going on in the tissues and organs of a man while he is actually alive, and accustoms himself to contemplate how these changes are disturbed, modified, or stopped in disease.

All our minute investigations upon tissues after death should be conducted for this end. The anatomical analysis should be carried out in the hope that the facts may be synthetically combined, and thus a mental image formed in which the actual phenomena of life are presented to the mind, and, as it were, seen in the perfection of their activity. The minute anatomy of healthy and morbid structures constitutes indeed the foundation upon which physiological and pathological knowledge in great measure rests. The facts of minute anatomy are, as it were, the dry bones which have to be afterwards clothed with active motor organs. The facts now learnt in the dissecting room and in the deadhouse, when contemplated by the trained and well-

stored mind of the thoughtful physician, are made to live. This is the result we are trying to accomplish. No work can be more worthy, and it will be to the advantage of mankind when more of the most gifted intellects are engaged in it.

As yet we are able to discern little of what is actually occurring in every part of the body during the life of man and the higher animals; but such imperfect glimpses as we have been able to obtain, have already thrown a flood of light even upon morbid phenomena which had hitherto seemed to be inscrutable and beyond investigation. Some idea of the progress that has been made, both in observation and philosophical generalization, may be formed by comparing the information we possess concerning the actual phenomena of "growth," "irritation," and "inflammation," with the fancies current but twenty years ago. We may feel sure that by further investigation we shall be able to realise more fully the actual changes proceeding in internal tissues and organs, although it may never be possible to submit these to actual examination during life. We may be prevented from obtaining anything but indirect knowledge, but nevertheless, the conclusions will, in some instances, be found to be not less reliable than others which are based upon actual observation.

In their essential nature many of the phenomena in the lower animals which can be seen and studied are like the corresponding phenomena in man, which cannot possibly be seen and studied during life; and hence we often resort to investigations upon the former in order to elucidate the nature of very important changes occurring in the latter. But even in man some of the most important of the vital phenomena may be witnessed.

He who has seen and well thought over the movements of a living white blood corpuscle, mucus or pus corpuscle, will be able to form a correct notion of many important changes which are continually occurring in various parts of the living organism and upon an extended scale. Very remarkable facts enable us conclusively to distinguish *living* from *non-living* matter, some of which may now be observed without difficulty under the microscope, with the aid of the highest powers, not only in many simple organisms but in man himself. There is no department of natural knowledge in which greater advance has been made than in this, which comprises investigations upon the nature of life. Facts which have been recently discovered enable us to draw a sharp and well-defined line between living things and the various forms of non-living matter, be it organic or inorganic, of simple or of complex composition. If, as investigation still further advances, the facts already known become confirmed, and the conclusions arrived at from recent researches supported by new observations and experiments, it will be generally admitted and taught that, in living matter, we have evidence of the operation of some agency, force, or power, which is distinct from every kind of physical force operating upon non-living matter. The

purely material doctrines so very popular only a short time ago, have already been modified in very important particulars.

The Microscopical Examination of Living Things.

135. Of Examining Living Bodies under the Microscope.—Some living things may be examined under the microscope without much difficulty. Many of the lower animals and certain vegetable tissues require only to be placed upon the glass slide in a drop of the water in which they live, and to be covered with thin glass, care being taken to prevent undue pressure. No cement or varnish must be used, but free access of air to the edges of the fluid must be permitted. In cases in which prolonged observations are required, arrangements, of which some will presently be described, must be made to compensate the loss of fluid by evaporation which would occur under ordinary circumstances. Again, many forms of living matter die when the temperature falls some degrees below the point at which their life is carried on. Hence, if such are to be studied for any length of time, we must have the means of uniformly maintaining the proper temperature, while the living matter is under observation in a glass cell under the microscope. Investigations of this character have been conducted with the greatest care during the last few years. Such success has been gained by the use of well-devised methods of research adopted by most skilful observers, that no doubt can be entertained that far more important results will be arrived at by new observers who shall devote themselves to the study of living matter, and still further improve the delicate instruments and apparatus required for conducting such researches.

136. Of Compensating Evaporation when Living Things are Examined in Water.—This may be effected, as recommended by Recklinghausen, by adapting a piece of sheet India-rubber tubing to the glass ring fixed on an ordinary glass slide, the diameter of the ring being sufficient to allow a piece of thin glass to be placed within its circumference. The upper end of the tube is tied round the object-glass of the microscope. Thus a "*moist chamber*" is made, and if one of Hartnack's "immersion lenses" be employed, observations may be continued upon a given object for some time. This form of moist chamber is, however, better adapted for use with low than with very high magnifying powers. Loss of fluid by evaporation may be compensated for if a little reservoir of water be fixed at one end of the slide, and a small piece of blotting-paper or silk thread be arranged so as to conduct the fluid to the object under the thin glass as fast as evaporation takes place. By placing a little cement round two-thirds of the thin glass cover, sufficient space is allowed for the requisite access of air, while at the same time loss by evaporation is reduced to the smallest amount. By this arrangement the growth of the spongioles of plants can be studied. Pl. XIII, fig. 8.

The most efficient method, however, that has been suggested for preventing evaporation of the fluid in which objects are being submitted to prolonged examination is that which has been very recently adopted by the Rev. W. Dallinger and Dr. Drysdale, in their researches into the life-history of monads. (See "Monthly Journal of Microscopical Science," March, 1874, p. 97.) The principle of this is to maintain sufficient evaporation from a surface of wet blotting-paper within the moist chamber to compensate for that which takes place from the edge of fluid exposed at the margin of the thin covering glass. The way in which this has been carried out is extremely simple and highly efficient. The same living object may be kept for days under a very high power. A ring of blotting-paper, which is kept continually moist, is arranged within the moist chamber, so that the included air is kept saturated with moisture. The details of construction are clearly described in the paper, and the apparatus can be constructed by the observer for himself.

137. Of Arrangements for testing the Influence of Gases upon Living Matter under Microscopical Observation.—A current of the gas contained in a reservoir may be directed upon the edge of the drop of water on the glass slide, but in this case we cannot be sure that its influence spreads very far beyond the point of its contact with the liquid. An afferent and efferent tube may be connected with the moist chamber described in the last section, the former being connected with the gas-holder. In this way more thorough exposure may be ensured, and the effects of different kinds of gases upon the living matter under observation may be studied.

138. Arrangement for passing Currents of Electricity through Living Bodies while under Observation.—Two platinum wires or two narrow strips of thin platinum foil may be cemented with marine glue to an ordinary glass slide at any desired distance apart. The other end is to extend beyond the ends of the glass slide, and to be so arranged that the wires from the battery or magneto-electric machine may be easily attached by the aid of a small clamp, or the metal ends may be prolonged into cups of mercury placed below the microscope on the table, and into which the terminal battery wires are made to dip. The object to be examined is placed between the terminal poles on the glass slide and covered with thin glass. When the object is in the field and properly focused contact is made, and the effect upon the organism or tissue studied. Kühne, Stricker, and many observers have studied the effects of currents of electricity upon various organisms while under microscopic investigation. Among the most instructive observations are those made upon the cornea of the recently killed frog and the muscles of the common maggot or larva of the blow-fly.—See p. 131.

139. The Warm Stage for maintaining Living Bodies at a uniform Temperature while under Observation.—In § 184, a very simple form of warm stage is referred to. The temperature of the heated air

can be regulated without difficulty by raising or lowering the wick of the lamp. The heated air comes into direct contact with the under surface of the slide, and its temperature can be ascertained by inserting a small thermometer into the heated chamber. This arrangement is susceptible of great improvement, but those who have conducted experiments more recently seem to have preferred hot water as the means of applying artificial heat to bodies under observation. The most satisfactory arrangement yet carried out is that adopted by Dr. Sanderson. It will be understood by reference to plate XIV, p. 148. The same apparatus may be employed for applying cold, and tubes for carrying gases to the chamber are also provided. This warm stage may be obtained of Mr. Hawksley, 4, Blenheim Street, Bond Street. See also § 184.

The student who determines to engage in this branch of investigation must be prepared for many difficulties of detail, the means of surmounting which will, however, occur to him if he continues to pursue his enquiries.

140. Of keeping certain forms of Bioplasm alive for Observation at Intervals.—A small quantity of pus, mucus, and fluids or semi-solid matters containing various kinds of living matter, may be preserved for some days without the death of the living matter they contain taking place by the following arrangement. A small glass tube about half an inch in diameter and an inch and a half in length is prepared, the edge of one extremity being turned outwards in the blow-pipe flame, so that very thin membrane may be tied over it. The tube is so arranged that the membrane just touches the surface of some distilled water in a small dish or capsule; the whole being placed in a moist hot-air oven maintained at a temperature of 100° F. In this way I was enabled to keep animal fluids freely exposed to the air, whilst evaporation was compensated for by the gradual imbibition of fluid from below through the pores of the membrane. I have succeeded in preserving mucus corpuscles and masses of living matter from the higher organisms alive for a considerable time longer than they would have lived at the ordinary temperature of the air. In this way one is enabled to imitate very closely the conditions under which changes go on in the body of the living animal. The plan is well adapted for a variety of observations upon the changes occurring in living things.

141. Of "feeding" Living Matter, and of the application of Chemical Re-agents to Living Bodies under the Microscope.—Many forms of living matter may be "fed" artificially with certain colouring matters in a very minute state of division. Thus, amœbæ will take up almost any foreign particles. White blood corpuscles, lymph and pus corpuscles, will take up particles of vermilion and other insoluble colouring matters, or the minute particles of carmine, which have been precipitated from an ammoniacal solution by the cautious addition of slight excess of acetic acid.

142. Of the use of Anæsthetics and Sedatives.—Various substances may be applied in solution to certain forms of living matter submitted to microscopical examination. Chloroform, ether, the salts of morphia, and other anæsthetics and sedatives, have been used for keeping minute organisms quiet while under microscopical examination. Dr. Maddox recommends a solution of from five to ten grains of hydrate of chloral to the ounce of distilled water. In examining worms, slugs, and insect larvæ of various kinds, this latter solution is said to be very useful.

143. Of studying the Movements of Living Beings.—Hitherto many of the movements occurring in living things have been referred to *contractility*, and, strange to say, the very authorities who never lose an opportunity of condemning those who attribute any phenomena in things living to the influence of a peculiar force or power—*vitality*,—consider that movements are sufficiently explained and accounted for if they are attributed to this so-called property of *contractility*. They do not attempt to define what they mean by the word, nor do they show in what this supposed property resembles or differs from other "properties" of matter.

If the student studies the question carefully, he will discover that great confusion has arisen from the circumstance that several essentially different kinds of movements have been attributed to this one property—*contractility*. Thus any tissue which alternately becomes shortened or lengthened, gaining in one diameter what it loses in another, is said to be contractile; while, on the other hand, that which moves in every conceivable direction is said to do so by virtue of this same property. It is not, however, very easy to see how two such different movements as repeated acts of contraction and relaxation within a definite space, and the actual movement of a mass of matter from place to place, can depend upon one and the same property. In fact, there are many different movements occurring in living things. A very little attention to the actual phenomena will indeed convince the observer that there are really different kinds of movements which are distinct from one another, and are due to very different causes. It is desirable to refer here more particularly to the movements taking place in a single elementary part or cell.

The movements occurring in living bodies may be arranged under the following heads:—

I. Primary or Vital Movements.—These *vital* movements have never been explained or accounted for. They affect matter only while it remains in the *living* state.

a. Cyclosis.—The circulation of the living bioplasm round and round a confined space or cell, as in many vegetable organisms.

b. Centripetal Movements.—By which the bioplasm gradually, and often but very slowly, extends from a point or centre, and spreads in various directions as nutrient matter is taken up by it, and changed and made to live. This is illustrated in the germination and growth of a sporule of a fungus. Every kind of growth and nutrition is dependent upon the movement of the particles of living matter.

c. Movements in all Directions.—Often occurring with wonderful rapidity, as in the amœba and the so-called amœboid organisms in warm weather. The slower movements of the bioplasm of the blood (white blood corpuscles), of mucus (mucus corpuscles), of pus (pus corpuscles), and of the cornea (corneal corpuscles), and other tissues.

II. SECONDARY MOVEMENTS indirectly occasioned by vital phenomena, but affecting matter in a non-living state.

d. Ciliary Movements.—Probably due to alterations in the quantity of fluid within the tissue. The changes in the proportion of fluid are probably brought about by the action of the bioplasm or living matter of the cell.

e. Muscular Movements.—Due to a disturbance (electrical or otherwise) in the neighbourhood of a contractile tissue—that is, a structure so disposed that its constituent particles are susceptible of certain temporary alterations in position, which alterations take place in certain definite directions only. The contractile tissue can only contract as long as fluids are made to flow through it by the agency of its living matter.

III. INORGANIC MOVEMENT not depending upon vital phenomena, but occurring in non-living as well as in living matter.

f. Molecular Movements.—Which affect all insoluble particles, *non-living* as well as *living*, if in a very minute state of division and suspended in a fluid not viscid.

g. Movements of Solid Particles suspended in Fluid, caused by Currents in the Fluid.—The pigmentary matter in the pigment cells of the frog. The motion being communicated to the particles by the fluid as it passes into, or out of the cell, through its permeable wall. This movement is dependent upon changes taking place external to the cell, and can be imitated artificially.

I.—VITAL MOVEMENTS.

Vital Movements are peculiar to matter in the living state. No movements like those to which I desire to restrict the term *vital* are known to occur in any matter which has not been derived from matter

in a living state. These remarkable movements cannot be explained by physics or chemistry, and are limited to living bioplasm. They have never been imitated. They cease when death occurs, and having once ceased cannot be caused to re-appear in the same particles of matter.

144. Cyclosis or Rotation of the contents of Cells.—The circulation in the cells of *vallisneria*, *anacharis*, *chara*, and *nitella*, *the hairs of the leaves of the common nettle*, *the purple or white hair-like appendages of the flower of the Virginian spider-wort*, may be observed without any difficulty. In all these the movement is due to the *vital* properties or powers of the bioplasm which moves round and round the cell; the hard cell-wall preventing its escape, and rendering movements in a right line impossible. The thinnest possible layer should be removed with a thin but very sharp knife, pl. VII, fig. 1, p. 78, from the surface of a young leaf of *vallisneria* or *anacharis*, and the two thin pieces thus obtained must be carefully placed on the slide with a drop of water and covered with the thinnest possible glass, care being taken to prevent the latter from pressing firmly upon the freshly cut surface. The movements are often sluggish, and sometimes completely cease immediately after the section is made. This is probably due to the shock produced in making the section. It is a good plan, especially in winter, to place the sections which have been made in water, in a small corked glass tube, which may be carried in the pocket for a quarter of an hour or more before they are to be subjected to examination.

Facts of the utmost general interest and importance may be demonstrated in vallisneria by the aid of the highest powers. The stream which moves round and round the cell, and looks like pure water under a twelfth, is found to be composed of extremely minute and apparently spherical particles of living matter, endowed with active motor power, if examined by a $\frac{1}{25}$ or $\frac{1}{50}$. The green chlorophyll masses are urged on by the actively moving living particles. One portion of the active, colourless, moving mass is seen to outstrip another portion, with which it gradually blends and incorporates itself, to be, in its turn, outstripped by other portions.

The hairs from the flower of the Virginian spider-wort (*Tradescantia Virginica*) are beautiful objects for studying the movements of the bioplasm or living matter in the cell. The transparent matter in active movement contains many minute highly refractive particles, which enable one to detect the slightest variation of the direction in which the stream sets. The young hairs of the nettle, the cuticular cells of this and many other plants, exhibit rotation. The movement can often be seen in the young, although it may not be visible in the fully formed cells. In some of the tender hairs of nettles grown in fern-cases, the movements of the bioplasm may be seen to perfection under a $\frac{1}{25}$ object-glass.

Solid particles are often suspended in moving bioplasm, and appear

to move of themselves, although, really, they are perfectly passive and are but carried in the moving stream. Sometimes these are formed from the germinal matter itself, sometimes they are foreign particles entering from without. The latter may be seen commonly enough in the amœba. Pus and mucus corpuscles and many other forms of living matter contain extremely minute particles, the nature of which has not been positively determined.

The bioplasm within the cells of desmids forms a transparent layer between the wall of the cell and the endochrome, which passes inwards towards the other part of the bioplasm called the nucleus. A clear spherical body, considered by many to be a *vacuole*, is often to be distinguished. This *vacuole* is not, however, a *space* filled with mere water, nor is it a cavity containing fluid within distinct walls. It is a spherical mass of pure, living, growing bioplasm—*a centre of life*, from and towards which minute particles of bioplasm and other matter, suspended in fluid, tend in constant streams. The solid particles suspended in the ever-moving currents, enable us to discern the movements of the semifluid bioplasm, which may be so very transparent and homogeneous that even under the highest powers it appears like water or perfectly pure colourless fluid. But that it moves is proved in many cases by the movement of solid particles suspended in it.*

145. Amœbæ can always be obtained by placing a small fragment of animal matter in a wine-glass full of common water and leaving it in a light part of the room for a few days. I have found it convenient to introduce a few filaments of the best cotton-wool into the water. The amœbæ collect amongst the fibres, which protect them from being crushed by the pressure of the thin glass when removed to the glass slide for examination. An imperfect idea may be formed of the changes taking place in the form of the most minute amœbæ by reference to fig. 1, pl. XII.

146. Colourless Blood Corpuscles may be conveniently obtained from the blood of many of the lower animals, or those from the blood of the human subject may be studied.

The white corpuscles in the circulating fluid of many mollusks and other animals exhibit vital movements, and are worthy of study. Such movements occur in every kind of living matter, but we are not able to demonstrate the movement in all cases. In the colourless blood corpuscle, however, and some other forms of bioplasm presently to be mentioned, they may be seen and watched without difficulty.

* The movements of the fluid with suspended globules in the vessels of the sheath of the bud of the India-rubber plant (*Ficus elastica*), which grows so freely in our dwelling-rooms, even in the midst of London smoke, may be well seen by placing a thin part of the sheath under an inch object-glass. Such a specimen is instructive, and it is well for an observer to study it before he attempts the demonstration of the more delicate phenomena of plant life under the higher magnifying powers.

In order to study the movements, a drop of blood or fluid containing the bioplasts is to be carefully covered with thin glass, so that a very thin layer may be obtained, but direct pressure must be prevented by inserting a piece of a hair or thin paper between the thin glass and the glass slide. Colourless blood corpuscles of the frog, toad, and newt, may be taken for studying the movements, which, however, are often very slow, particularly in winter, and sometimes no movement may be observed for some minutes after the corpuscles have been under observation. The movements of the white blood corpuscle of the human subject are sometimes very remarkable, and are sometimes wonderfully active in some of the corpuscles. Upon what the variation in the degree of activity of the movement depends is not known. In the year 1863, I described the changes which might be seen to occur under a very high power, and gave a figure of a corpuscle from which many elongated processes of bioplasm had extended some distance from the general mass of the corpuscle, but processes of every conceivable form may be seen in many instances, pl. XII, fig. 5.

The colourless corpuscles of the blood differ in age, and if a very thin layer of blood be examined with care under a $\frac{1}{25}$ or $\frac{1}{50}$, corpuscles differing much in size and also in transparency will be discerned. Some of the smallest are less than the $\frac{1}{100000}$ of an inch in diameter, but I believe that the matter of which they are composed is the same as that which constitutes the basis substance of the ordinary white corpuscles. Indeed I think many of these very minute particles of bioplasm in the blood which I described and figured in 1863, might probably grow into ordinary white blood corpuscles and take the place of those of the latter which had been transformed.

After white blood corpuscles and other bioplasts have been allowed to remain still for some time, a very transparent circular or spherical space appears in some part of the matter. This has been termed a "vacuole," and it has been supposed to be a space filled with transparent fluid like water. But really this perfectly clear waterlike matter is bioplasm that still remains alive after much of that constituting the corpuscle has died.

It is inaccurate to call these white corpuscles and other bodies *leucocytes*, because they are neither white nor cell-like. They are masses of bioplasm, and facts in connection with their movements go far to prove that they are destitute of any cell-wall. It is not correct to call them *white* corpuscles, but the term has got into such general use that nothing would be gained by objecting to it now. As regards such a word as *leucocyte*, however, there is an affectation of precision which is very objectionable on many grounds. Many terms recently introduced are more vague than the old ones they replace.

The colourless blood corpuscles have been termed *contractile cells*,

and *amœbiform corpuscles*, as if all things which manifested this peculiar property of movement were of an *amœbiform* type. I have shown that the lymph and chyle corpuscles, the white blood corpuscles, the so-called mucus and pus corpuscles, young epithelial cells, &c., are but free masses of living bioplasm, which being destitute of any firm cell-wall, and embedded in a more or less fluid medium, are free to move in every direction. In short, the movement in question is characteristic of not only the *amœba* and the so-called *amœbiform* cells, but of every kind of living matter, although it cannot be seen in every instance, in consequence of the slow rate at which the changes in form occur, or other circumstances.

At the present time we are acquainted with so many cases in which active movements may be actually seen—and every year new examples are discovered—that we are justified in concluding that all bioplasm possesses this property. The changes in relative position and alterations in form which take place during development in the case of the bioplasm of the firmest tissues, prove that this form of living matter is no exception. The movements of the amœba must therefore be regarded as *vital movements*, not peculiar to it and a few other things. The terms *amœbiform* movements, and *amœbiform* bodies, should no longer be employed, since *spontaneous movement*, *growth*, *multiplication*, and *formation*, are attributes of every kind of living matter, but are not manifested by any kind of non-living matter that is known.

147. Mucus Corpuscles are embedded in the mucus which is found upon the surface of the mucous membrane of the nares, fauces, and air tubes. By coughing sharply or by making a violent effort—something between coughing and sneezing—small portions of transparent mucus may often be detached. The transparent viscid mass is placed upon a glass slide, and covered very carefully with a piece of the thinnest glass, which is to be very gradually but not too firmly pressed down, so as to cause the mucus to spread out, and form a very thin layer. The movements occurring in a mucus corpuscle are represented in pl. XII, fig. 2.

148. Pus Corpuscles should be obtained from a mucous or other surface at the time that they are *growing and multiplying*. Pus, which is usually examined, consists of *dead*, not of *living corpuscles*. These are *spherical*, as generally represented in books, and many have a sharp, well-defined outline, owing to coagulation having occurred upon the surface. It is in this way the so-called membrane or cell-wall of the pus corpuscle has resulted. A cell membrane may always be formed artificially, by exposing the surface of a mass of albuminous material to the influence of a re-agent, or to conditions which are known to effect the coagulation of albumen.

The best specimens of pus for studying vital movements may be

obtained from the urine in some cases of *chronic inflammation of the bladder*. Not uncommonly in this affection the urine contains very little solid matter, and the pus corpuscles retain their vitality, although immersed in it for many hours after the urine has been removed from the bladder. Indeed in some instances the corpuscles retain their vitality for days. So far from the outline of the corpuscles being circular, as usually figured and described, in many specimens not a single corpuscle of this form is to be detected. I have examined specimens of pus from urine in which every corpuscle exhibited little "buds," "offsets," or "protrusions" at every part of its circumference. Examples are given in pl. XII, fig. 6, and the attentive examination of such specimens, even under moderate magnifying powers, will convince the observer that the corpuscles are slowly undergoing alterations in form. The movements are very remarkable. Movements occur in the most minute of these buds or offsets, which have been detached as represented in pl. XII, fig. 3. In fig. 8 a drawing of some of the particles from vaccine lymph has been introduced.

One of the smallest of the particles of living matter detached from a pus corpuscle is capable of absorbing nutrient material and growing into a corpuscle, having all the properties and powers of that from which it was derived. If the student will only examine into these things he will soon be able to picture to himself the wonderful changes which occur in inflammation, and in the multiplication of the living virus of contagious fevers.

In order to watch the vital movements above referred to, it is only necessary to place a very small portion of the pus on a glass slide, and cover it very carefully with a piece of the thinnest glass, pressure upon the corpuscles being prevented by the introduction of a portion of hair or a small piece of tissue paper or thin writing paper as already described. In very cold weather it is desirable to warm the slide slightly, or to use one of the arrangements referred to in § 139.

149. Bioplasm Cells from the aqueous humour of the Frog's Eye, certain Corpuscles in the connective Tissue, and young epithelial Cells from the Mucous Surfaces generally, exhibit vital movements of the same kind. Masses of bioplasm in the frog's cornea, especially when under the influence of a current of electricity, exhibit active movements (Kühne). See § 138.

150. Ova.—Vital movements are manifested by the bioplasm of which the germinating part of all ova is constituted. Much may be learnt by patiently studying the changes taking place from day to day in the ova of the common water snail (Limnæus stagnalis) and other mollusks. As is well known, the fertilised ova of many amphibia, reptiles, fishes, and mammalia, are the seat of wonderful movements and changes in form. The movement of the mass in some cases commences before,

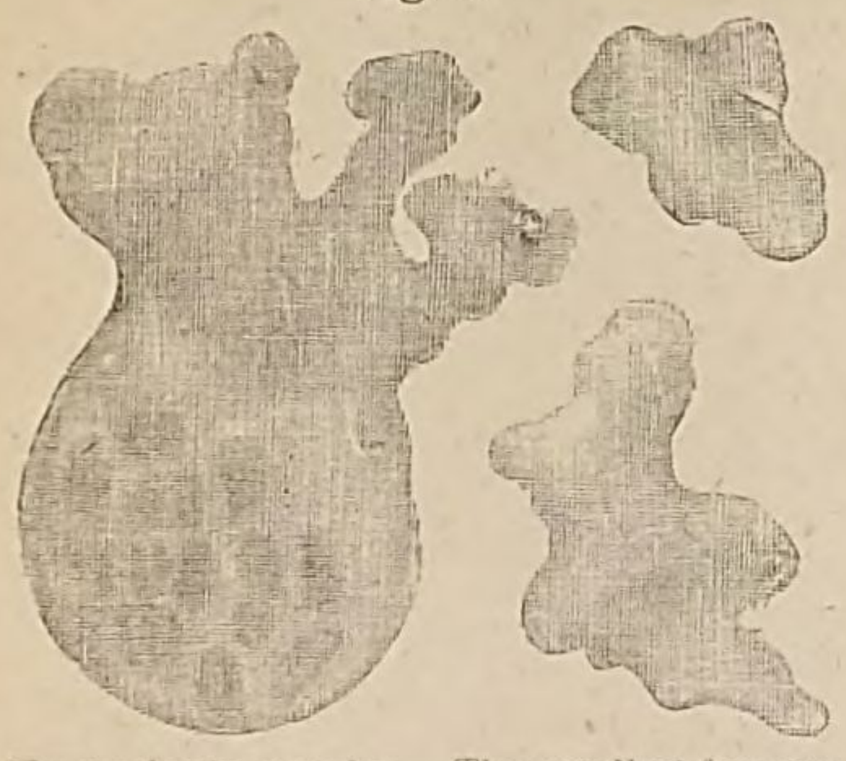
and always continues for a long time after, impregnation has been effected. The result of the movements is the concentration of the germ yolk at one part of the surface, and the division of the entire ovum into this and the food yolk. It is probable that the layer of bioplasm around the food yolk is the seat of the active movements and not the yolk itself. That part which is known as the germ yolk is alone the seat of all those wonderful phenomena which result in the formation of the embryo. The phenomena may be well observed, and with great facility, in the ova of frogs, newts and many osseous fishes.

Dr. Ransom, of Nottingham, who has for some years paid the greatest attention to the examination of the ova of fishes, recommends those of the *Stickleback* and the *Pike* as well adapted for observation. It is not necessary that the ova of the latter fish should be fertilized even. No form of active living matter is better adapted to the purposes of the physiologist than that of the egg of the pike. In making experiments upon either of these fishes, Dr. Ransom divides the spinal cord just behind the edge of the gill covers, and then the fish is amenable to manipulation, and yet lives and breathes well. In order to impregnate the ova of the stickleback, the same observer recommends that the ripe male should be cut open and his testes used in fragments, as the semen cannot be well *squeezed* out. The application of heat and cold in investigations of this kind has been referred to in § 139.

151. Nature of the Movements of Bioplasm.—The movements in bioplasm can be distinctly seen with a twelfth of an inch object-glass. It is often necessary to examine the bioplasm very attentively for half a minute, for in some cases the changes in form are so slow that the observer who looks at the object carelessly cannot satisfy himself of the actual occurrence of movement at all. It is useless to attempt to conduct observations of this kind in a careless, off-hand manner. Those who desire to have the delight of pondering over such changes will gladly find the leisure to observe the facts. This is, however, just one of those phenomena which, having been well seen once, can generally be detected afterwards without difficulty. Under the sixteenth, twenty-fifth, or fiftieth, the alterations in form can be observed very clearly. There are few things more wonderful, or which will furnish more interesting matter for careful thought and for valuable and useful speculation than the appearances seen.

No satisfactory explanation of the movements has yet been offered, and though I know many scientific men will attribute them to "antecedent changes" in the moving matter, and rest content—refusing to discuss the nature of the antecedent changes they imagine—it seems to me that by doing so they fall into the fatal mistake of acting in a manner indicating a desire on their part to check the progress of scientific enquiry. It looks as if they feared enquiry might extend beyond the exact point at which

Fig. 1.



Very minute amœbæ. The smallest free particles exhibiting very active movements were less than the ten-thousandth of an inch in diameter. × 5000. p. 124.

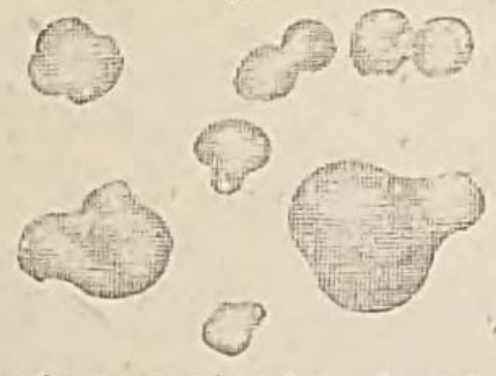
Fig. 2.

Mucus-corpuscle. From the mucus of the throat of man in health; showing the different forms assumed by the living mass within a minute. p. 124 1863. × 2500.

Fig. 3.

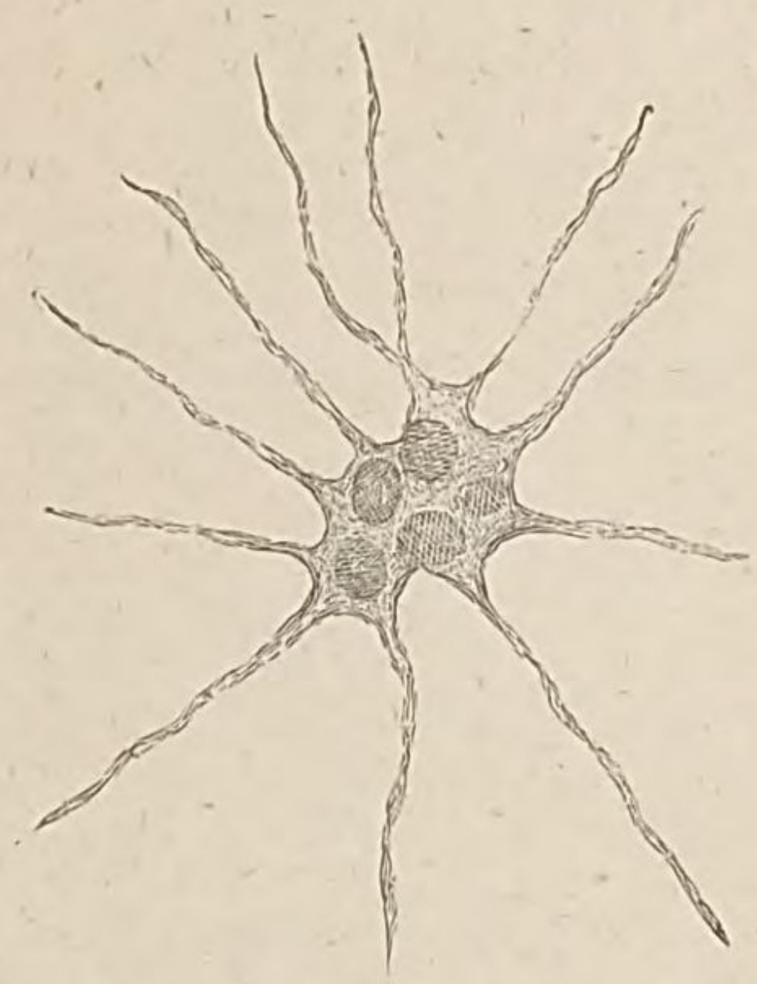
Different forms assumed by the same minute mass of bioplasm of pus from the bladder during five seconds × 2800. p. 125.

Fig. 4.



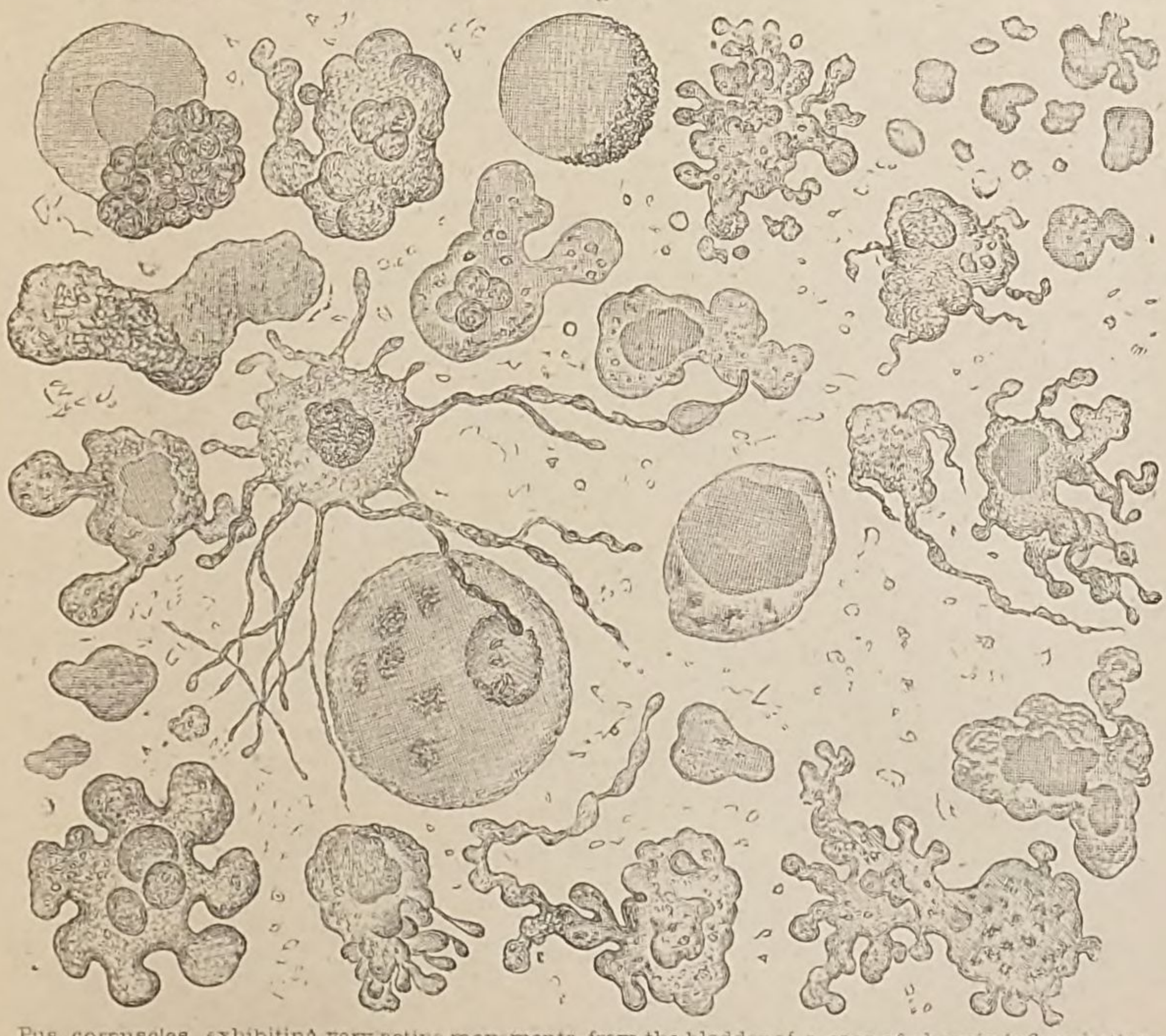
Particles from vaccine lymph, exhibiting very active movement × 5000. p. 125.

Fig. 5.



Changes in form from movements of bioplasm. Colourless corpuscle, an hour after being drawn from finger. p. 123. × 1800.

Fig. 6.



Pus corpuscles, exhibiting very active movements, from the bladder of a case of chronic inflammation. Man. Showing alterations in form due to vital movements. × 1300. p. 125.

[To face page 126

it must be stopped if the interests of those who teach narrow physico-chemical doctrines to which so many scientific authorities have prematurely committed themselves, in direct opposition to all the teachings of true science, are to be considered paramount.

The movements which have been characterised as *vital movements* in the sections of this chapter, I regard as *primary*, and think that the power of movement exists in connection with the matter of which each small portion of the moving mass is composed. It will appear to many minds unsatisfactory to attribute the movement to the influence of a power of the nature of which nothing is known; but it is surely more reasonable to adopt this view, which may turn out to be correct, than to assert these movements are due to physical force, when it is well known that they cannot be explained by any laws. An unprejudiced person who thoroughly studies the movements and carefully thinks over the facts of the case will, I think, find himself compelled to admit that they cannot be adequately accounted for in the present state of knowledge, unless the existence of some sort of supra-physical or *vital power* be assumed. No one would be more ready to acknowledge that these movements and other phenomena characteristic of living matter were due to ordinary energy than I should be if the correctness of such a doctrine had been proved, or if any approach had been made towards this end; but as a working physiologist, desiring to promote to the utmost real advance in this department of science, I consider it a duty to oppose as strongly as I can the practice pursued by many scientific authorities in the present day, and especially in this country, of reiterating the assertion that *all*, or *nearly all* the phenomena of living beings are to be accounted for by the action of ordinary physical forces, while it is a fact that not one of the phenomena peculiar to living beings can be thus explained. Instead of the objections raised to such assertions being answered, or the challenge to consider the matter in detail being accepted, we are authoritatively told that the "tendency of modern science" is towards this apparently much desired end, and that although living matter cannot yet be prepared by man, "the day is not far distant when its artificial production will be rendered possible," and so forth!* Now this is prophetic dogmatizing, not science.

Every student ought to endeavour to detect the fallacies which underlie many of the modern doctrines, and especially those in the writings of very self-confident philosophers, and which fallacies are often so cleverly concealed by the ingenious choice of words, that they are not easily discovered, except by one who has accustomed himself to analyse modern dogmatic assertion. It has been stated, with what is

* See "Protoplasm," 3rd edition. "The Mystery of Life." "Life Theories and Religious Thought." "Bioplasm."

mistaken for learned precision, that force is "conditioned" by the "*molecular machinery*" existing in the cell, but no doubt it has been concluded by those who made the assertion, that no one would be very likely to enquire what the "molecular machinery" was like, and whether it was to be seen, and how the "conditioning" took place. It is a fact that the changes in question occur without the existence of anything to which the term *machinery* can be properly applied. Instead of the living cell being like a machine, it is perhaps less like a machine than anything else of which we have any knowledge. This "living *machine*" is just a very minute mass of soft, colourless, transparent, semifluid matter, endowed with very wonderful properties or powers,—in which matter is decomposed and its elements re-arranged, while forces are conditioned in a manner that cannot be effected by man with the aid of the most perfect machinery and elaborate apparatus his ingenuity has devised. Living matter cannot then be a machine. It does not act upon the principles of any machine, nor is force conditioned in it as it is in a machine,—nor have the movements occurring in living matter been explained by physics, or the changes which take place in its composition by chemistry. The phenomena occurring in living matter are peculiar, differing from all other phenomena; and therefore, until we can explain them, they may be fairly distinguished by the term *vital*. Not the slightest step has yet been made towards the production of matter possessing the properties which distinguish living matter from matter in every other known state. As investigation advances the movements of living matter, or bioplasm, will acquire a still higher importance; for there can be no doubt that the alteration in the position of elementary parts during the formation of tissues is due to vital movements of the bioplasm of which the "cells" in their early stage consist.

II.—Secondary Movements.

The secondary movements to be referred to, are indirectly due to vital phenomena; and though they occur in *tissue* and not in *bioplasm*, must, to a certain extent at least, be referred to *vitality*. Tissue cannot be alive in the sense that bioplasm is alive; for it does not *grow*, neither can it *select*, or *form*, or *nourish itself* as bioplasm is known to do. The movements occurring in the tissue or formed material are indirectly brought about by bioplasm. They cease when the bioplasm dies, and they could not be induced if no living matter was present. The material which is the seat of these secondary movements is peculiar, and its existence is a consequence of the vital properties of the bioplasm which took part in its formation. Bioplasm, therefore, contributes to the movements in question in two ways:—It takes part in forming the matter that

moves, and it is instrumental in producing the *changes* which result in movement.

152. Ciliary Movements.—Ciliary action is, I think, due to changes going on within the elementary part or "cell." It probably depends upon currents which flow to and from the bioplasm or living matter, and the altered tension which is thereby caused in the formed material or tissue of the cilium. Ciliary motion cannot be dependent upon nervous action, and clearly it is not to be explained by any disturbance in the surrounding medium. Ciliary motion ought not, I think, to be regarded as a *vital* movement, although due to changes which are consequent upon vital phenomena. Cilia consist of "formed material."

In the immediate vicinity of ciliated cells, as for instance, in the mouth of the common field snake and the boa-constrictor, are sometimes observed cells with open mouths, out of which mucus and various substances, formed or secreted in the interior of the cell, pass. In the formation of these products, nutrient matter from the blood, after passing through the attached extremity of the cell, is absorbed by the living matter. At the same time the outermost portion of the bioplasm becomes converted into the peculiar contents of the cell, and thus the formed matter which has been already produced becomes pushed towards the orifice. Both classes of cells result from bioplasm. In the one bioplasm seems to be converted into mucus which escapes, in the other into processes of formed material—cilia—which grow from the cell.

Cilia of Infusoria.—Different forms of ciliary action may be observed among the different species of infusoria. It is, however, doubtful if many of the very fine spine-like bodies, the movements of which seem under voluntary control, can be correctly regarded as cilia. The simple organisms of this class seem to possess the power of permitting or stopping the vibrations, but there can be no doubt that in vertebrate animals generally, ciliary action is quite independent of volition. There is certainly no connection between the cells of ciliated epithelium and the nerves. The movement of many hair-like appendages have been included under the head of "ciliary action," although they are not of this nature.

Cilia of Frog's Tongue. Branchiæ of Oyster and Mussel.—Cells of ciliated epithelium in active vibration can always be obtained by scraping the back of the frog's tongue. In the mucus which is removed, numerous cells will be found. The thin glass cover must be prevented from pressing too firmly by inserting a small piece of thin paper beneath it. The student may also obtain very beautiful ciliated particles of epithelium in active vibration from the branchiæ (gills) of the oyster or mussel. Some of the cilia from the latter situation are of considerable length, and occasionally the vibration of a single cilium may be studied, and the observer may demonstrate the interesting fact that movement occurs not only at the base of the cilium, but in every part of the vibratile filament.

Cilia in Uriniferous Tubes of the Newt's Kidney.—Of all the ciliated structures known to me, the newt's kidney is, I think, the most beautiful and the most remarkable. The tortuous uriniferous tubes in the upper thin portion of the kidney are lined in their whole length with ciliated epithelium, which continues in active motion for some time after the removal of the organ from the body of the animal. In order to display this wonderful object, we must proceed as follows :—After destroying the newt by cutting off the head, the abdominal cavity is to be laid open, and by turning the viscera to one or other side, the kidneys may be exposed. Towards the pelvis of the animal, the kidney presents much the same appearance as it does in the frog : but, upon tracing it upwards, it will be found to become gradually thinner, and to extend quite into the thoracic portion of the animal. It is this *upper thin part of the kidney* which shows the ciliary motion to the greatest advantage. See pl. XIII, fig. 2. A probe, *a*, is represented in the drawing underneath that portion of the kidney which should be examined in the microscope. The secreting tubes lie upon one plane, so that a single tube may often be seen in the field of the microscope at one time. In every part of the highly convoluted tube from the Malpighian body downwards, the most active ciliary movement is to be seen. Many solid particles are often observed in the current of fluid which is urged downwards with such velocity. A more beautiful object under a half-inch object-glass, can scarcely be conceived. The thin portion of the kidney, above referred to, is to be very carefully removed from the body by delicate manipulation with fine forceps and a pair of scissors, moistened with a little water, or, what is still better, with some of the serum of the animal, placed in a large thin glass cell, and carefully covered with thin glass. The cell should be sufficiently thick to prevent any pressure upon the preparation. After the ciliary action has ceased, the cilia are with great difficulty distinguished. I have frequently noticed that many of the tubes in the lower thick part of the kidney do not exhibit ciliary action, and believe this is due to the circumstance that many of the old tubes are undergoing degeneration, while new ones are being developed, and those advancing towards maturity. I have been able to find in some newts tubes in every stage of wasting.

Air Tubes of Man.—Ciliated epithelium may also frequently be obtained from the larynx and trachea of man by coughing violently. The vibration will continue for some time after the specimen has been transferred to the glass slide, especially if it be gently warmed. The observer will be surprised at the enormous number of cilia projecting from a single cell from the human trachea ; indeed it often happens that a mass is expelled which seems to consist of hundreds of long filaments all in active vibration, radiating from a common point.

153. Contractility of Muscle.—Contractile tissues are characterised

by a repetition of movements. *Contractility* is essentially different from any form of *vital* movement The first affects various kinds of formed material only; the last is peculiar to bioplasm. Vital movement is continuous. Contractility is interrupted. By vital movements matter may be raised, particle over particle, higher and higher. Contraction alternates with relaxation. It is, as it were, a vibration to and fro—the alternate shortening and lengthening of a fibre within a given space. *Vital movement* may occur in a mass of living matter in any direction. *Contraction* takes place in one definite direction only, and never alters.

Contractile movements may be watched in many of the lower animals. The alternate contraction and relaxation of the spiral stem of a vorticella is a beautiful object of study, but the tissue is perfectly transparent and structureless. Muscular contraction may be studied in any of the small insects or crustacea. Mr. Bowman strongly recommends muscular fibres from a young crab (Phil. Trans., 1841). Some of the large muscular fibres from the leg or other part of the large water beetle (*Dytiscus marginalis*) are favourable for studying muscular contraction. Many small transparent aquatic larvæ are also advantageous for the examination of muscular fibre during contraction.

The phenomena of muscular contractility may, however, be studied more satisfactorily in the broad muscles just beneath the skin of the common maggot or larva of the blow-fly, than in the muscles of any other animal I am acquainted with, and as these can be readily obtained at any period of the year, I recommend them for observation. The movements, which are very beautiful, continue for ten minutes or a quarter of an hour after the muscles have been removed from the body of the recently killed animal, so that a specimen may be prepared and passed round the lecture room in one of the portable microscopes, § 2. In the winter I have seen the contractions continue for upwards of half an hour. But the most instructive method of examination is under the influence of polarised light, with a plate of selenite. When the ground is green, the waves of contraction which pass along each muscular fibre in various directions, are of a bright purple. In other parts of the field the complementary colours are reversed. There are few microscopic objects, that I am acquainted with, so beautiful as this. With the aid of very high powers, the actual change occurring in the contractile tissue as it passes from a state of relaxation to contraction, and from this to relaxation again, may be studied, and for many minutes at a time. In order to obtain the muscular fibres of the maggot, it is only necessary to slit up the larva, and after removing the viscera, to separate some of the muscles from the outer skin to which they are attached. They may be moistened with some white of egg, saliva, or better than all, a little of the colourless fluid from the animal.

III.—Inorganic Movement—Communicated Movements.

154. Of Molecular Movements.—When any solid matter in an exceedingly minute state of division is suspended in a limpid fluid, every one of the minute particles is seen to be in a state of active motion or vibration in the neighbourhood of other particles. Molecular movements may be studied in the case of any matter in a very minutely divided state, suspended in water. Charcoal powder, phosphate or carbonate of lime or magnesia, Indian ink or gamboge rubbed up with a drop of water, or even a thin layer of milk will afford an illustration of molecular movements. The cause of these movements has not yet been fully ascertained, and they have often been mistaken for *vital movements*. If some *bacteria* developed in any decomposing water be exposed to a temperature of 200°, they are destroyed, but although quite dead, *molecular movements* still occur. If, however, the movements of the dead particles be compared with those of living bacteria, a great difference in character will be noticed.

155. Movements of Granules within Cells.—The movement of insoluble particles from one part of a cell to another, as occurs in the radiating pigment-cells of batrachia, is probably due to alteration in the direction of the flow of fluid in the cells,—*from* the cavity of the cell *towards* the tissues, or *from* the surrounding tissue *into* the cell. If the capillaries were fully distended, fluid would permeate their walls and would pass into the cavity of the cells, in which case the insoluble particles would gradually become diffused and would pass into all parts of the cell ; while, on the other hand, if the capillaries were reduced in diameter, and the lateral pressure upon their walls diminished, there would be, as is well known, a tendency for the fluid in the surrounding tissue to flow towards the vessels and pass into their interior, pl. XIII, fig. 4. In this case the quantity of fluid in the cell would be gradually reduced, and the insoluble particles would become aggregated together, and would collect in those situations where there was most space, as in the central part of the cell around the nucleus. Moreover, in the last case, the flow of fluid, which constantly sets towards the nucleus, would be instrumental in drawing the particles in this same direction, while if the cell contained a considerable proportion of fluid, the currents would pass between the particles without moving them. Evaporation, as it occurs after death, of course causes concentration of the insoluble particles towards the centre of the cells.

These changes in the pigment-cells of the frog have, however, been considered by Professor Lister to be due to *vital actions*, and he agrees with Wittich and others who maintain they are under the immediate control of the nervous system. Indirectly no doubt they are, but I do not think that any experiments have proved satisfactorily that the nerves exert any *direct* influence upon the movements of the particles in these

cells. It is well known that the nerves govern the calibre of the vessels, and they thus indirectly influence the amount of fluid in the surrounding tissues. In this indirect manner only can nerves be said to affect the movements of the particles in the cells. The reader will find a full account of Professor Lister's experiments, and the arguments deduced from them, in his paper, "On the Cutaneous Pigmentary System of the Frog," published in the Philosophical Transactions for 1858.

Of the Phenomena of the Circulation.—Action of the Heart and Arteries during Life.

156. Movement of Blood: of the Heart's action.—Movement of the Blood.—For examining the circulation in the web of the frog's foot, a young frog with a thin web should be selected. The body and one hind leg are loosely bound up in wet rags, the other leg being allowed to protrude. The body is then tied to the frog-plate, and fixed in the proper position for observation of the circulation of the blood in the vessels of one of the webs. The flow in a small artery, in a vein and in the capillaries must be separately studied. A drop of plain water, or water with $\frac{1}{4}$ per cent. of common salt is to be added before the web is covered with thin glass.

The frog-plate is a piece of brass-plate with numerous holes at the sides for tying the animal carefully wrapped up in wet rag in a convenient position in order to prevent struggling while under observation. At one end of the plate a piece of glass is let in, and across this one of the webs is stretched; strings previously attached to two of the toes are passed through the proper holes so as to display the web advantageously. A frog-plate may be easily made of thin wood, or a piece of gutta-percha.

By careful observation of the circulation, first of all under a low power, and then under a quarter of an inch object glass, most important and highly interesting facts will be learnt, and if some irritant be applied to one part of the web, the early changes occurring in inflammation may be demonstrated. *See* p. 139.

Of observing changes in the Circulation.—In cases in which it is necessary to conduct observations on the circulation with the aid of high powers it will be found desirable in practice to increase the length of the tube of the microscope, instead of employing object glasses of very high magnifying power. A quarter of an inch object glass may thus be made to magnify as highly as a twelfth, and as the distance between the object glass and the thin glass covering the web is very considerable, there is not the same danger of serious derangement every time the animal moves slightly, as when the glass of the object glass nearly touches the thin glass. Several different lengths of tube may be adapted to the microscope body, which may be thus increased to the length of two feet or more, if desired.

If a small artery be brought into focus and the tip of one of the toes be very lightly touched, the artery is seen to contract immediately, but somewhat irregularly in different parts of its course. Sometimes a few blood-corpuscles are firmly compressed, and for several seconds the vessel remains so strongly contracted that not a corpuscle passes along it. By performing this instructive experiment, the observer may form a notion of the wonderful contractile power of the coats of the smaller arteries, and demonstrate conclusively that the afferent nerve fibres distributed to the skin of the foot generally, influence the nerve centres from which the nerves ramifying amongst the muscular fibres of the arterial coats take their rise. This is a beautiful instance of reflex nervous action affecting the vessels.

The lungs of the frog and newt have been submitted to microscopical examination, and the mesentery of the same animals is advantageous for studying the capillary circulation. The circulation may also be studied during life in the capillaries of the tail of a small fish, minnow, stickleback, carp, &c. The fish should be wrapped up in wet lint and loosely tied at one end of a glass slide, the tail being placed about the centre, and covered with a piece of very thin glass.

Action of the Heart.—A more correct idea of the action of the heart may be formed by watching its contractions in a small living animal under the microscope than in any other way with which I am acquainted. A small fresh-water mollusk such as the planorbis, often shows the movement of the heart very beautifully, and as the organ consists of a single auricle and ventricle only, it is very favourable for study. A young fish, or newt, or frog tadpole may be taken for the purpose, but I have found that a young snake removed from the egg exhibits the phenomena most beautifully. The blood may be distinctly seen as it eddies through the various apertures of the fœtal snake's heart in passing to or from the different vessels and cavities. The undulating contractions of the auricles and ventricle of the heart are very wonderful. Under a two-inch power adapted to a binocular microscope, a far more correct idea of the heart's action will be formed than can be obtained by reading the best descriptions that have been given by authors.

The branchiæ of the frog tadpole or young newt may be examined in a flat glass cell specially prepared for the purpose, and by an arrangement of tubes the animal may be supplied with fresh water while it remains under observation. In pl. XIII, fig. 1, p. 144, is represented a form of cell, which I made some years ago for a proteus, but a cell for a newt or other animal may be made upon the same plan.—H. to W., § 131. The circulation of the blood in the capillary vessels of a mammalian animal may be studied in the thin membrane forming the "wing" of a young bat.

157. Chyle.—For studying the movements of the chyle in the lacteals, a mouse, rat, or young rabbit may be taken. The animal should be fed

with a little lard beaten up with a piece of pancreas and a small quantity of bile, so as to form a soft pultaceous mass which may be strained through muslin. About half an ounce, or less, of the cream-like fluid may be injected by the aid of a small syringe into a flexible catheter which has been passed down the gullet into the animal's stomach. After a couple of hours, the creature should be pithed, stunned, or destroyed very suddenly, and a small portion of the mesentery with the intestine attached withdrawn through an aperture in the abdominal walls and submitted to microscopical examination with a low power.

The examination of all the moving objects alluded to in this section, may with advantage be conducted with the aid of the binocular. The circulation in the cells of vallisneria, and the movements of the cilia of small animalcules or ciliated cells under a high power with the new binocular of Messrs. Powell and Lealand, once seen can never be forgotten, for the mind seems to realize the actual state of things occurring during life, in a manner which before was not possible.—*See* p. 22.

Phenomena of Growth and of the Multiplication of Living Particles.

158. Of Growth and Multiplication.—The observer who aims at studying the remarkable and highly interesting phenomena of germination, growth, and multiplication of the bioplasm of cells or elementary parts, in the tissues and organs of man in health and disease, will find it advantageous first to investigate these processes in the simplest living beings where they occur under conditions less complex. He must exercise the utmost caution in drawing inferences from what he sees, or rather thinks he sees, and he must always bear in mind that great and irreconcilable differences of opinion exist among even distinguished observers with regard to the general nature of the changes which take place when, for example, a spore of common mildew germinates, or an insignificant bacterium gives rise to new bacteria. How, therefore, is it likely that the mode of growth, origin, and multiplication of some of the highly complex structures formed in man, especially in the course of disease can be described with correctness or fully explained to the student?

The observer will learn many most important facts by watching the germination of the common mildew, and studying the different appearances of the plant when developed under different circumstances. It is exceedingly instructive to watch the growth of the spongioles of a young plant (mustard, wheat, mignonette, or those of any very small seed), as they grow under the thin glass. Fluid may be constantly supplied according to the plan described in page 116.

I know no example of the growth and development of vegetable tissues more instructive than is to be found in some of the large-leaved begonias, now common greenhouse plants. Buds will form on the petiole

and leaf ribs, if the former be inserted into moist sand and kept in a warm place for a week or two. It has been said that the buds are transformed hairs, but this is by no means invariably the case, for I have seen the bud developed beneath the epidermis in a situation in which no hair existed. The development of the minute leaves in the microscopic bud by the division of cells, and the orderly arrangement and growth of the resulting particles will excite interest. Sections may be made through buds at different stages of development and preserved in glycerine without difficulty.

The mode of origin and multiplication of a bacterium and the growth of a spongiole of a plant, may appear to be questions far indeed removed from the province of medical enquiry, and yet we shall find that by such investigations only can we hope to determine the nature of some phenomena, the true explanation of which lies at the very root of a knowledge of the real nature of disease. To gain any real advance, either in physiology or in medicine, we must establish certain fundamental truths; and unless we are content to examine and revise again and again the first principles of our science, we cannot hope to progress. Let not the student of medicine, therefore, conclude that the multiplication and growth of the lower forms of animal and vegetable life are not in his province. There is, indeed, scarcely a department of natural knowledge which does not bear more or less directly upon medicine, and those who discourage careful research, or speak disparagingly of its results, are doing their utmost, unconsciously it may be, and perhaps unintentionally, but nevertheless most effectively, to retard progress.

Since the greatest physician cannot yet give a satisfactory account of what is going on in an ordinary cold, or the greatest surgeon explain to us the phenomena which are connected with the formation of a common boil, it is surely time that those inclined to study these things should be permitted to do so without having their hard struggling life ruffled by the scoffs of people who are over-proud of having discovered some short and easy path to popularity, but along which no earnest, thoughtful student would care to make his way.

Abnormal Increase.—Multiplication of Bioplasm.—Inflammation.

159. The Formation of Pus.—Inflammation consists essentially in the growth and multiplication of the bioplasm of a tissue or organ with unusual rapidity. Bioplasts which in health may slowly increase in size, and divide and subdivide, grow perhaps ten or twenty times as fast as they should grow, and may produce as many descendants in twenty-four hours as, in the normal state, would have resulted in many weeks or months.

The abnormal bioplasts thus produced, have *gained* as regards their rate of growth and multiplication, but they have deteriorated in formative power, if indeed they have not altogether lost it, and it is in formative power that the bioplasm of a tissue differs from the degraded forms of living matter.

It has been generally admitted that in inflammation, cell-multiplication, or "proliferation," takes place, but attention has not been drawn to the fact that it is *the bioplasm only of the cell* which really takes part in the "proliferating" process. The cell wall, where it exists, as well as every kind of formed material, is perfectly passive; and although it has been affirmed over and over again that in those instances in which the cell exhibits a constriction the formed material has "grown in," and that when a cell divides, the segmentation is due to the active in-growth of the formed material, it is certain that the change occurs in a very different manner. The proportion of the tissue will, in some cases, account for the physiological and pathological changes in the part, but to the bioplasm only all the active phenomena of life must be referred, *Growth* without bioplasm is impossible, and *inflammation* in the absence of bioplasm is not conceivable. Far-fetched theories out of number have been invented in order to explain upon physical principles the fact of the increase and subdivision of a cell, but one after the other they have been discarded. In order to account for the apparent subdivision of the nucleus, that is of the bioplasm, in cases of inflammation, it has been gravely suggested that pus-corpuscles made their way from the deep aspect of the tissue through the cell wall, and in this way gained the interior of the cell. White blood-corpuscles or lymph cells have been conceived as wandering out of their ordinary places and making their way into merely conjectural cells. Such ideas are unnecessary and superfluous. Every degree of enlargement and subdivision of the bioplasm of cells in inflammation may be seen by examining many specimens of cells from an inflamed mucus surface such as the vagina, the urethra, or the bronchial tubes. In 1861 I pointed out that the increase of the bioplasm in the epithelial cells of the cuticle could be very successfully demonstrated after the application of a blister, and every stage of change from the increase of the normal bioplasm up to the formation of the so-called "inflammatory product," pus, was clearly made out. The examination was conducted under magnifying powers of from 700 to 1,500.

The formation of pus in vaginal epithelium may be studied in the deposit of the urine from any case of leucorrhœa. The formation of pus corpuscles by the growth and division of the bioplasm of these cells is very distinctly proved, and illustrative specimens may be easily prepared.

The student will, however, obtain more conclusive evidence of the gradual changes which take place during the progress of the inflammatory process in such a texture as cuticle in inflammation. If a

small blister be applied to the arm every stage of change from healthy cuticle to pus may be studied. When the blister has properly risen, a piece of the upraised cuticle may be snipped off, and its under surface carefully examined. The changes of the bioplasm may be demonstrated more distinctly in specimens which have been carefully stained with carmine, but they are evident enough in fresh specimens examined in some of the serum of the blister. The bioplasm of the inflamed epithelial cells will be found to be much larger than that of the corresponding normal cuticular cells. The bioplasm of the deeper cuticular cells will be found to have increased in greater proportion than the rest—in some instances to such an extent as to project beyond the thin layer of formed material of the cell. Such projecting portions are often detached, and the free particles grow rapidly in the exudation poured out from the blood-vessels. They divide and subdivide. Thus is formed the great majority of the free bioplasts which exist in such number in relation with the under service of the upraised cuticle of the blister. Some of them, however, no doubt result from the growth of minute particles of bioplasm derived from the blood and suspended in the fluid which transudes through the walls of the capillaries as I described in a paper published in the Quarterly Journal of Microscopical Science, 1863. That all the free bioplasts in pus were once white blood-corpuscles, each one having passed through the capillary wall, and then insinuated itself through the chinks between the deep cells of epithelium, as has been recently assumed by many,—is not only a conjectural proposition, but one that may be proved erroneous by any one who examines proper specimens for himself, and it is only needful to ask the reader to contemplate the facts that are assumed, to be sure that he will form a correct judgment. Let him consider the time occupied in the occurrence of the changes described, the number of the supposed colourless corpuscles that have changed their place, the distance each one must have traversed in its course from the blood to the situation in which it is supposed to have assumed the condition of a pus-corpuscle. The hypothesis is one of many which are the delight of ingenious speculators, and which minds deficient in inventive power are only too anxious to support and spread as if they had been well-ascertained truths resting upon observation and experiment.

160. Vascular Phenomena of Inflammation.—The process of inflammation as it occurs in man and vertebrata is complex, and is made up of phenomena which may be arranged in two classes :—

1. The growth and multiplication of the bioplasm already considered in the last section.

2. Changes in the vessels.

The *vascular phenomena* may be well studied in the foot of the frog, arranged as described in § 153, but the mesentery and the tongue are used in preference by some observers. The changes have usually been studied

with powers such as the half inch and quarter magnifying under 250 diameters. This amplifying power is, however, too low to enable us to make out some important points. By the arrangement described in page 133, the power of the quarter is easily increased without any of the inconveniences which are experienced in using very high objectives for the examination of such a specimen. If, however, young animals be used, a twelfth, magnifying 700 diameters, may be employed without difficulty. The ordinary frog-plate may be used or a special one prepared, a ring of cork being arranged around the hole so that the part to be studied may be properly fixed with the aid of very fine pins, while the animal is kept perfectly still by the use of anæsthetics or by subcutaneously injecting a $\frac{1}{5}$ per cent. solution of curari.

Inflammation may be excited by passing a fine thread through the part, or by constriction with a ligature, by making a cut in the tissue or removing a small piece, or by the application of chemical irritants. A fragment of Cayenne pepper or a small piece of mustard, or a very small piece of the material of which "Rigollot's mustard-leaves" are made, a little strong acid, or a hot wire, may be used to excite inflammation. A small vein from an inflamed frog's foot is seen in fig. 3, pl. XIII.

The demonstration is by no means so easy as would be inferred from the descriptions that have been given; and although it is quite certain that the corpuscles do pass through the walls of the vessels, it is doubtful if the phenomenon is either a constant or necessary part of the inflammatory process as it occurs in man and vertebrata as has been maintained, —while it is quite certain that pus-corpuscles are formed by the enlargement and division of tissue bioplasts (see Part II), as well as by the growth and multiplication of white blood-corpuscles. The view that all pus-corpuscles are colourless blood-corpuscles which have made their way through the vascular walls is, I think, untenable.

The migration of the colourless blood corpuscles upon which so much has been written, and which was first seen by Addison and Waller, may be studied in cold-blooded vertebrata.

Observations on inflammation as it occurs in warm-blooded animals are much more difficult, but Stricker, with the aid of the warm stage, appears to have been able to study some of the changes. The bat's wing, the mesentery and the cornea of small animals are the parts usually selected, but the arrangements to be successful require such care and skill, and the practical investigation presents so many difficulties that it should not be attempted by the student until he is well accustomed to delicate investigations of the kind.

I strongly recommend the student to study specimens of inflamed tissues which have been prepared after death in the manner I have described in Chapter VII, and I feel sure that if the facts demonstrated by such specimens were carefully compared with those demonstrated

upon the living animal, most valuable conclusions would be arrived at from the two methods of study conjointly.

The outlines of the so-called epithelium of the vessels may be demonstrated by injecting a $\frac{1}{4}$ or $\frac{1}{2}$ per cent. solution of nitrate of silver. The vessels after injection are to be well washed in water in the dark, then exposed to light, and examined after having been soaked in glycerine in which medium the specimens may be preserved permanently. The reader will find much information in connection with this department of enquiry in Prof. Sanderson's Handbook.

Aeroscopy.

It has long been known that great numbers of solid particles composed of inorganic matter, and of organic matter dead and living, are suspended in the air at all times and at different elevations. As obviously must be the case, the suspended particles have been found to vary much in character and in numbers at different elevations, at different periods, and under different circumstances. Great nonsense has been talked and written on this subject, and the most absurd "discoveries" have been made both in air and water, and paraded before the public with the only effect of exciting amusement and ridicule. Air contains in suspension besides organic and inorganic particles varying according to the character of the dust raised upon earth, numerous living organisms, minute ova, and germs of various kinds, some of which are in an active, while some are in a quiescent state. Pasteur obtained many sporules of fungi from the atmosphere even at considerable heights. These, when transferred to a suitable medium, he found would grow and multiply.*

161. Of Solid Particles suspended in Air.—The various particles suspended in the atmosphere may be very easily obtained for microscopical examination. The "dust" which has accumulated upon any shelf, will supply the observer with a great variety of highly interesting and important objects. The dust usually contains particles of carbon, starch granules, scales of moths, fibres of wood, fragments of cotton, flax, wool, hair, and feathers, sporules of fungi of various kinds, epithelium from the skin, and many other bodies according to the place or district in which the examination is made. The particles may be collected at any time by exposing a shallow dish containing distilled water, to the atmosphere which it is desired to examine for foreign particles. A more effectual method is to place some ice in a perfectly clean glass vessel, and allow the moisture of the air to condense upon

* Pasteur's solution for growing fungi is composed of the following ingredients :—

Sugar, 10 grammes.
Tartrate of ammonia, 5 decigrammes.
Yeast ash, 1 ,,
Water, 100 cubic centimetres.

the outer surface. The drops of water should be collected in a suitable vessel placed beneath. The condensed fluid will of course contain any insoluble particles which were suspended in the air. The water is placed in a perfectly clean conical glass, to the lower part of which the insoluble particles held in suspension will gradually subside. They may then be removed with the aid of a pipette, placed upon a glass slide, and submitted to microscopical examination under the highest powers.

A better plan is to draw air through a tube, the interior of which may be moistened with a little perfectly pure glycerine, or to cause the air to impinge upon the surface of a glass slide moistened with the same substance. The necessary current of air is easily produced by connecting a glass tube with a reservoir of water, so arranged that when the water is allowed to flow out, air from the apartment must be gradually drawn through the tube. Many modifications of this arrangement will occur to the mind of the observer. He may put in practice the plan which seems to him best adapted for the particular investigation he is about to undertake.

Air containing living particles may be collected in closed vessels, and if retained for a short time the living particles sometimes grow and multiply. The living particles though too minute or too few in number to be detected, may be caused to germinate. Different kinds, and the same kind in very varying numbers, are obtained at different heights and under varying conditions. If it is desired to prevent the particles from germinating, it is only necessary to add a little carbolic acid. A one or two per cent. solution is strong enough.

Bibulous paper, or perfectly clean glass plates moistened with glycerine, may be suspended in the air of an apartment to catch the solid particles suspended in it. By proceeding in this way, however, the observer will be overwhelmed with the vast number of the particles he will see; and, as may be supposed, it is extremely difficult to identify bodies, which although very similar in appearance, may be essentially different in their nature. Minute living particles of pus or other living matter in a state of active vitality, although capable of giving rise to the most terrible and fatal diseases if they obtained entrance into the living organism, would be very readily passed over, or, being obscured by the mass of foreign matter present, could not be seen. It is said that Dundas Thompson detected such particles in cholera wards, so long ago as 1849 and 1854, and Eiselt subsequently discovered pus in the atmosphere of an ophthalmic ward.

My own researches have demonstrated how pus-corpuscles and other masses of bioplasm form buds or offsets, and divide and subdivide, minute offsets often becoming detached. Such minute particles would be readily supported by the atmosphere. I have also shown that in

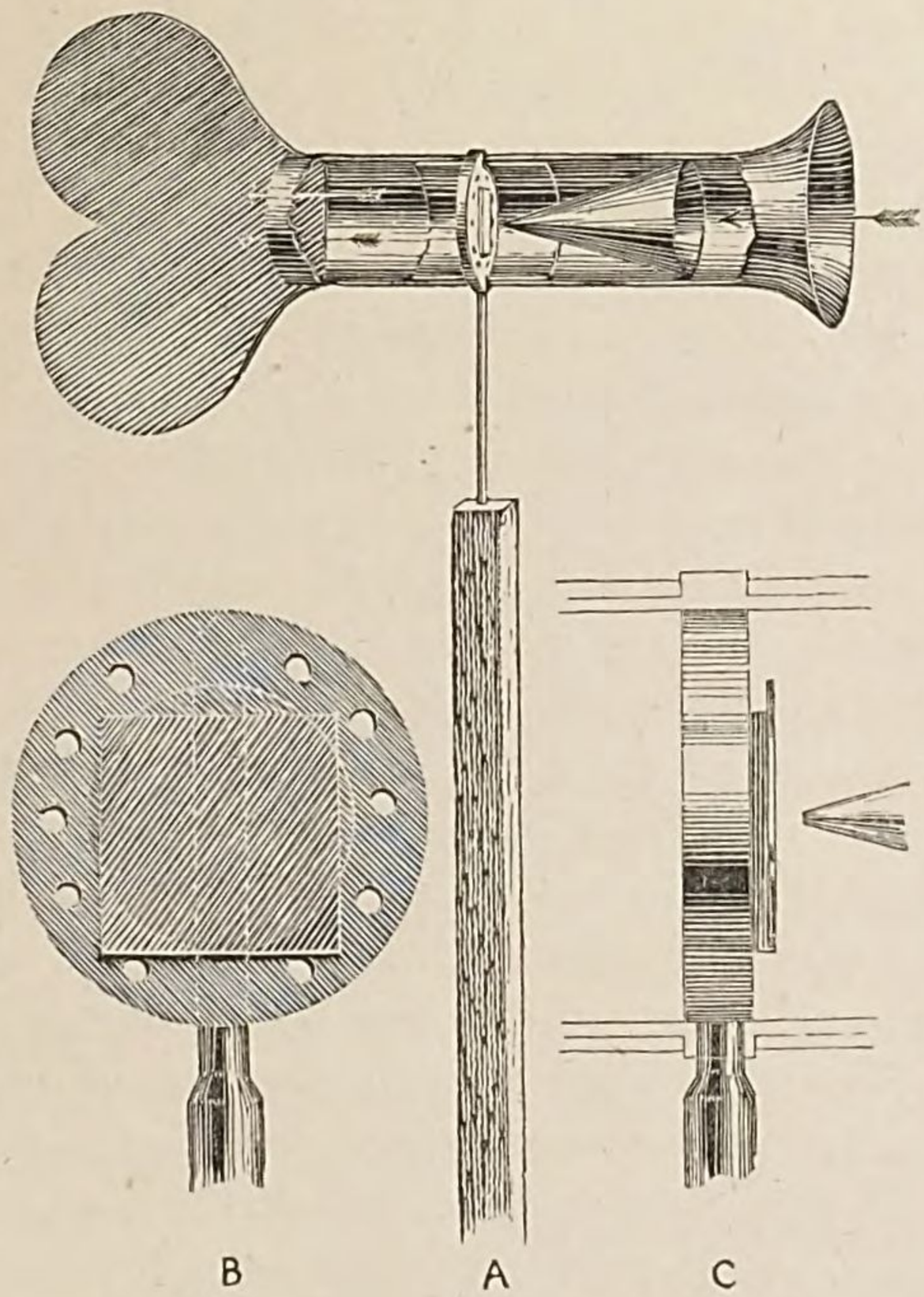
contagious diseases, besides the particles of altered epithelium, pus, and allied forms of bioplasm, are other living particles exceedingly minute, which perhaps alone possess contagious properties. These would very easily pass from one organism to another. (See my Report to the Cattle Plague Commissioners, 1866.) In the preparations examined by Dr. Cunningham in India, "minute monad-like molecules and globules of an undefined nature, abounded." Similar descriptions have been given by other observers who have worked in England.

This is a branch of advanced and extremely delicate microscopical enquiry, which we have as yet scarcely entered upon. Though surrounded with great difficulties, the investigation will probably yield the most remarkable and important results, and it is most desirable that it should be pursued by many observers. It must be borne in mind that the very high powers recently made by Messrs. Powell and Lealand are likely to be extremely useful in examining living bodies suspended in the atmosphere, and there is even reason to think that we may be able to discover characters in some of these bodies by which they may be recognised again. The small microscope devised by Prof. Brown is admirably adapted for researches of this kind. See p. 24.

162. Aeroscope.—An excellent form of aeroscope has been devised by Dr. Maddox. It consists of a well poised vane so arranged that even when there is but a very slight wind it will be turned, and a current of air will enter the wide end of a funnel-shaped tube, and impinge upon a glass slide moistened with glycerine arranged opposite its narrow extremity leaving the solid particles, while the air escapes at the side. I have described this arrangement in my work on "Disease Germs," p. 161, and have given figures taken from Dr. Maddox's paper published in the Monthly Microscopical Journal for June 1, 1870, p. 286. When the instrument is to be used in perfectly still air, a draught through the apparatus is caused by adapting a long tube through which a current of heated air ascends as long as a spirit or other lamp is kept burning at its lower extremity.

Dr. Cunningham has recently improved upon Dr. Maddox's vane. The arrangement is seen in the accompanying figure copied from Dr. Cunningham's work "On the Microscopic Examination of Air."—(Sanitary Commission of India, published in Calcutta). Preparatory to taking observations, "the apparatus" which is made of brass tubing, was well washed with spirits of wine and heated over a spirit-lamp. A microscope cover-glass of suitable size was then carefully cleaned, and one surface smeared with pure glycerine. A minute drop of the same medium was placed upon the diaphragm-plate, and the *dry surface* of the cover-glass applied to it, leaving the smeared surface exposed. By the glycerine on the diaphragm the glass was secured in a vertical position, and in this way the necessity for a spring was avoided, "the

use of which was found inconvenient." The particles that had collected were examined when the cover-glass had been in a position for twenty-four hours or longer. Some of the sporules deposited upon the slide will have undergone development by the time the microscopical examination is made. If the slide be kept in a warm place, and the glycerine in which the particles have been collected be much diluted with water, most of the living particles will have germinated, but this, if desired, can be prevented by the addition to the glycerine of a little pure carbolic acid.



Apparatus employed by Dr. Cunningham, in India, for collecting atmospheric dust for microscopical examination. *a*, vane; *b*, diaphragm-plate enlarged; *c*, sectional view of collecting glass shown on a larger scale.

163. Pollen Grains in Air.—Mr. Charles Blackley of Manchester adopted several plans for collecting pollen grains from the air for the purpose of microscopical examination, in his investigations upon hay fever. A current of air was made to impinge against a glass moistened with a solution consisting of water, one part, glycerine one part, proof spirit two parts, with five grains of pure carbolic acid dissolved in each ounce of the solution. Mr. Blackley, however, found that invariably a relatively larger number of grains were deposited during short periods of time. Ultimately a simple plan recommended by Dr. Phœbus, Dr. Salisbury, and others was adopted. Several glass slides each having a space of one centimeter square were coated with the fluid above referred to. These were carefully placed upon a little stage surmounting a post and protected by a cover placed a few inches above. After exposure for twenty-four hours, each slip was examined under the microscope, and the number of grains in the measured space carefully counted.

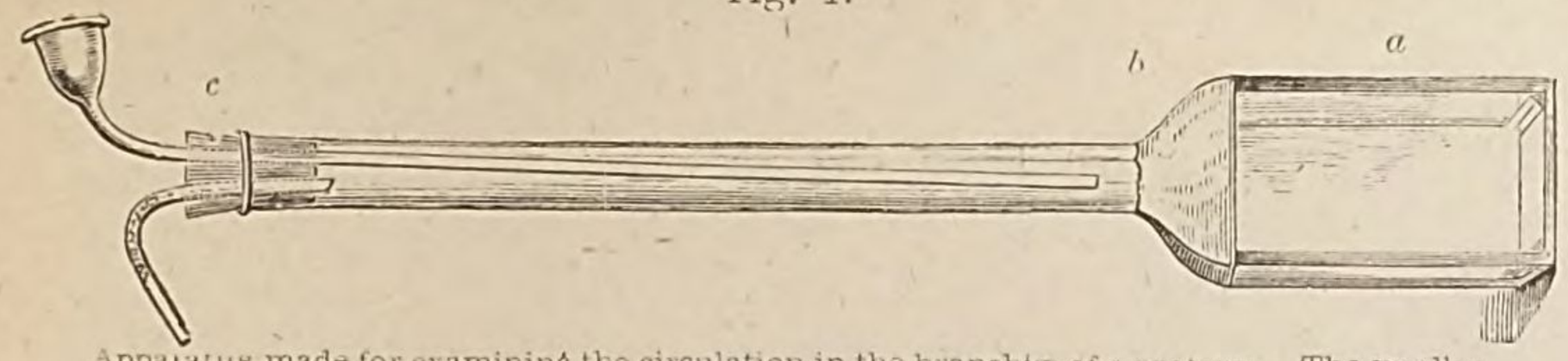
Mr. Blackley found that the number of pollen grains increased from the beginning till the end of June. More grains were found when the air was dry than in damp weather. He also discovered that the higher strata of the air were often richer in pollen grains than currents near the earth.* Mr. Tichborne has shown that air taken at a great height excited fermentation at least as readily as air taken from an overcrowded dwelling.†

164. Bacteria germs in Air.—Dr. Sanderson has been led to conclude from his experiments that bacteria do not exist in the air, but other observers profess to have seen these bodies among the particles separated from the atmosphere, or at any rate, they have seen bacteria which if not in the atmosphere must have been developed from germs that were in the air. It must be borne in mind that an actually living growing bacterium is a living body which contains a very large proportion of water. A particle of the kind could hardly be free in the air, for it would almost immediately become attached to one of the many floating particles which are many thousands of times larger than itself. In a dry atmosphere the living bacterium would soon shrivel up, and its life would be destroyed. Actively growing and multiplying bacteria pass very quickly through the several phases of their existence and die. We should, therefore, not expect to find moist living bacteria in great numbers on the slide placed to catch the suspended solid particles by the aeroscope. But on the other hand the germs of bacteria may exist in countless multitudes in the atmosphere, deposited in and upon the insoluble particles of various kinds which are always present. These under favourable circumstances will germinate, and the resulting bacteria grow and multiply. In a very moist atmosphere living bacteria would probably be found. While then it is probably perfectly true that bacteria are generally propagated in liquid media, and that very few, if any, exist ordinarily in the atmosphere, it may not be quite accurate to assert that no bacteria germs are suspended in the air or exist in connection with *any* particles floating in the atmosphere. Besides, germs exist which are so minute as to be perfectly invisible when first formed, even under the highest magnifying powers yet made. They grow, however; and, after a time, become large enough to be distinctly visible. It must also be borne in mind with reference to several statements that have been made concerning the destruction of bacteria by heat, that an active growing multiplying bacterium is killed at a temperature far below that to which a bacterium germ may be exposed, and for a considerable time, without its vitality being destroyed. Ordinary growing bacteria are destroyed even by desiccation at ordinary temperatures, but no one

* "Experimental Researches on Hay-Fever or Hay-Asthma." By C. H. Blackley, M.R.C.S.

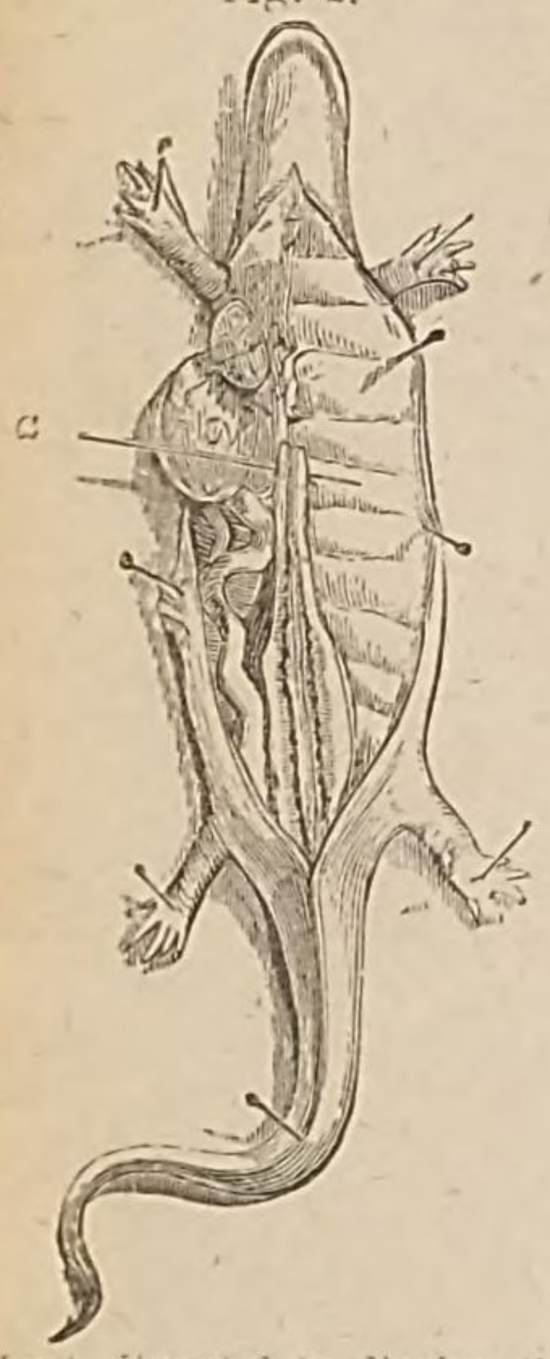
† "Chemical News," October, 1870.

Fig. 1.



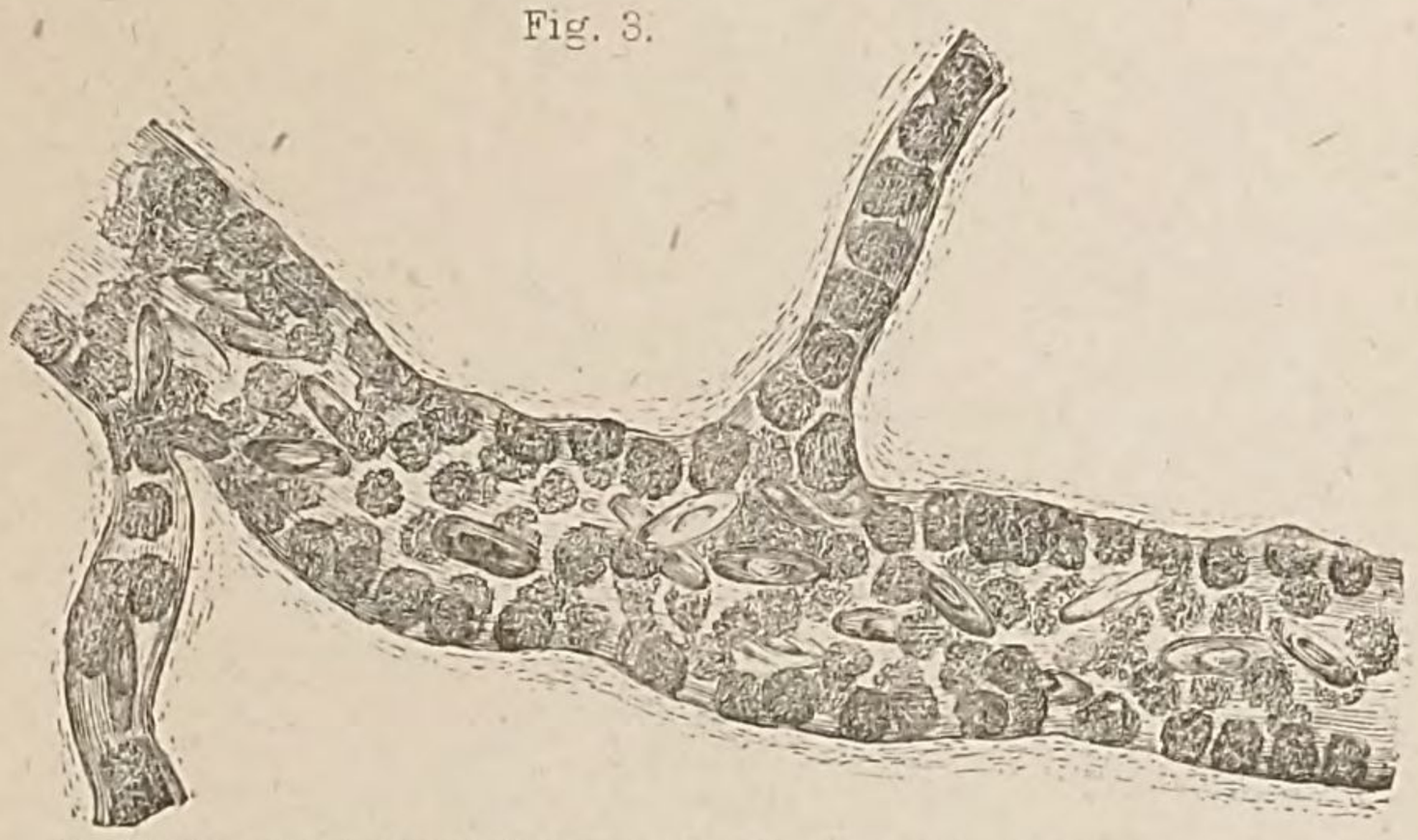
Apparatus made for examining the circulation in the branchiæ of a proteus. The smaller tubes were for the purpose of supplying the animal with fresh water. p. 131.

Fig. 2.



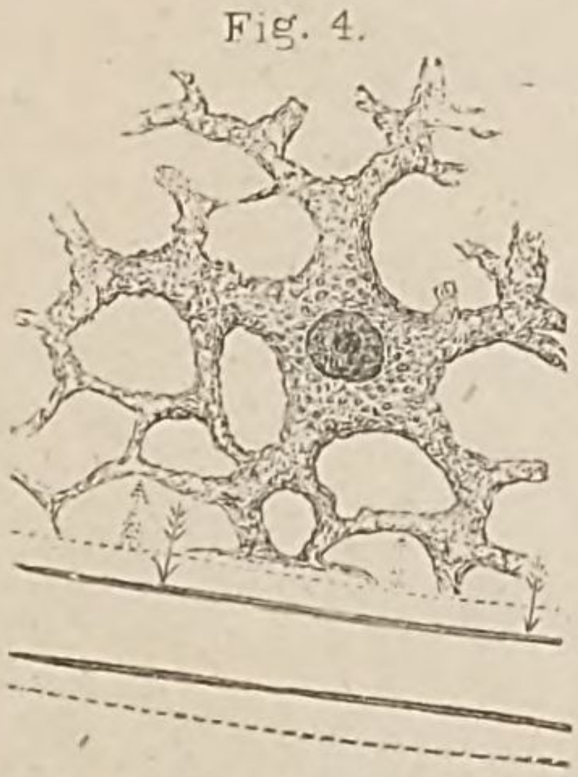
Newt dissected to display the thin part of the kidneys. p. 130.

Fig. 3.



Small vein in web of foot of very young frog, a few minutes after inflammation had been excited by mustard. × 215. p. 139.

Fig. 4.



Pigment cell of the frog and its relation to a capillary; showing direction of flow of fluid in different states of contraction of the vessel. p. 132.

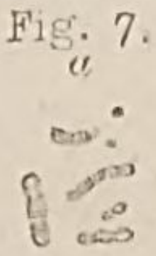
Fig. 5.

Most minute bacteria. × 2800.

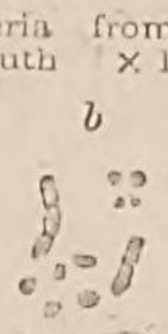
Fig. 6.

Bacteria undergoing germination. × 1800. p. 115.

Fig. 7.



Bacteria from the mouth × 1800.



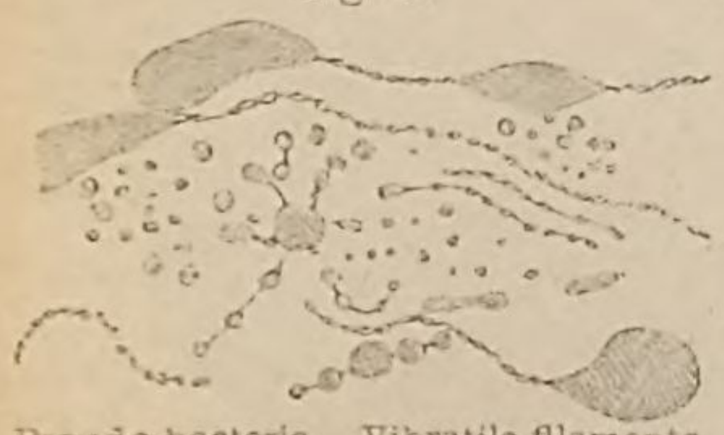
The same in old epithelial cells of the mouth. × 3000.

The same × 5000. p. 146.

Fig. 8.

Simple arrangement for conducting water to living bodies under observation. p. 116.

Fig. 9.



Pseudo-bacteria. Vibratile filaments and minute particles, consisting of viscid coloured matter. Blood corpuscles, human subject, after being subjected to heat. × 1800. p. 145.

Fig. 10.

Curious spontaneous changes in form of red blood corpuscles of the frog. × 700. p. 145.

[To face page 144.

has yet succeeded in proving that bacteria *germs* are destroyed by this process, even though they be kept in any ordinary dry place for years.

165. Bacteria germs not Contagious Poison.—The interest attached to bacteria is due to the circumstance that they have been regarded as the cause of contagious disease. The contagium or poison of contagious diseases is, however, not of this nature. Bacteria will live upon those forms of morbid bioplasm which constitute contagia just as they will appropriate the matters resulting from the death of normal bioplasm or from the decomposition or decay of organic fluids and solids. An individual living growing bacterium probably lives for a very short time, but it is rapidly succeeded by others. As has been stated the actively multiplying bacteria are much more easily destroyed than the germs (*micrococci* or *mycrozymes*) from which they spring. It has been supposed by some that bacteria originate spontaneously, but of the so-called experimental facts hitherto advanced in favour of this doctrine of spontaneous generation, and of the inferences deduced, many cannot be considered convincing. Not a few of the observations have been, so to say, read the wrong way, for they may be accounted for more satisfactorily without resorting to any such far-fetched hypothesis as abiogenesis. A case may be made in these days by clever discussion for almost any doctrine that may be put forward. No one can prove that the higher animals did not originate spontaneously; but it seems clear that the arguments yet advanced in favour of the spontaneous origin of a bacterium are no better than those that might be adduced to support the idea of the spontaneous origin of a worm or a dog. Futile will be the determined and repeated efforts to force people to believe that these ever-present, growing and multiplying bacteria are actually *disease germs*.* The evidence is unsatisfactory, and many of the statements untrustworthy. I believe that never before in the history of science have been made such violent efforts to foist upon the mind doctrines that never had anything to recommend them as of late years. Some distinguished persons are now always trying to make us accept certain scientific views which are contrary to evidence. The question of the real nature and origin of disease germs will be again referred to in Part II, and further remarks upon the fungus theory of contagious disease offered.

166. Of Pseudo-Bacteria.—It has been stated over and over again that bacteria originate in decomposing matters, and one who has recently written on the subject thinks that he has *seen* the fibrillæ of muscle resolve themselves into these living bodies! It is always necessary to be on our guard against such fallacious observations. Those who have had much experience in the manufacture of pseudo-bacteria could produce a number of objects and advance facts and

* See "Disease Germs," 2nd edition, 1873.

arguments which would probably fully convince any inexperienced person that there was abundant evidence to prove that bacteria were but the modified particles of certain tissues. But in truth the evidence points entirely the other way. Perfect looking bacteria may be produced readily enough by gently warming over a spirit lamp a little blood placed on a glass slide and covered with thin glass. From the red blood corpuscles under these circumstances numerous, very narrow-jointed filamentous processes are seen to project, and from their constant vibration and molecular movements these might be easily mistaken for living bodies (pl. XIII, figs. 9 and 10). Sometimes they become detached and move about in a manner much resembling certain forms of bacteria. At the same time any one familiar with investigations of this kind would not be deceived either by the general appearance or by the movements of these bodies. True bacteria are represented in pl. XIII, figs. 5 to 8.

167. Of Detecting insoluble living or lifeless Particles in the Breath.—It is very easy to collect solid particles from the breath. By causing each successive expiration to come into contact, during a minute or two, with the surface of a piece of clean glass, the object in view may be attained, but by breathing through a glass chamber kept cool by the application of ice externally, a greater number of insoluble particles may be collected in the condensed moisture. The observer will be surprised at the number and size of the particles which are suspended in the expired air ; oil globules, epithelial cells, and portions of mucus corpuscles (?) may sometimes be detected.

In examining the breath of cows, I was surprised to find large fragments of the food, starch corpuscles, several different kinds of very dark, irregular, insoluble particles somewhat resembling those of soot suspended in the air of London rooms, three or more different kinds of fungi sporules, bacteria, fragments of epithelium, and a number of bodies I was unable to identify. These all collected upon a clean plate of glass moistened with pure glycerine which was held for a few minutes in the current of the animal's breath.

Another method of obtaining insoluble organic and inorganic particles suspended in the breath, is to place some perfectly clean cotton wool in a glass tube which is so connected with a mouthpiece, that the air must pass through. Any solid particles are obstructed by the wool. The air is, as it were, filtered by this process. The wool may be examined dry or after having been moistened with a little weak glycerine. This plan was very successfully employed by Mr. Crookes in his experiments upon Cattle Plague. He found that the poison or virus suspended in the breath of a diseased animal was obstructed by the cotton wool, a fact which was demonstrated by inoculating a sound animal with the wool which had been thus exposed. The animal re-

ceived the disease. Mr. Crookes sent me portions of wool for examination, and upon carefully comparing the exposed wool with some of the same wool which had not been so exposed, differences were observed under the highest magnifying powers. Minute particles were detected in the former which could not be found in the latter. I think it very likely that if such experiments were conducted with extreme care, some most valuable facts might be discovered concerning contagious diseases. I hope that ere long in one or two instances of eminently contagious diseases in the lower animals we may be able to catch and preserve the living particles of *contagium* on their way from the infected to the sound organism. Gun-cotton has been recommended, and it has been suggested that after exposure it might be dissolved in ether, and thus the particles obtained perfectly free, but I should think this process scarcely likely to yield reliable results. The great difficulty in all such experiments is to obtain the particles for experiment sufficiently clean and perfectly free from foreign particles. Still there is no doubt that if this matter be carefully studied, many new and valuable methods of investigation will soon be discovered, and highly important facts bearing upon that most interesting question, the nature of contagion, demonstrated.

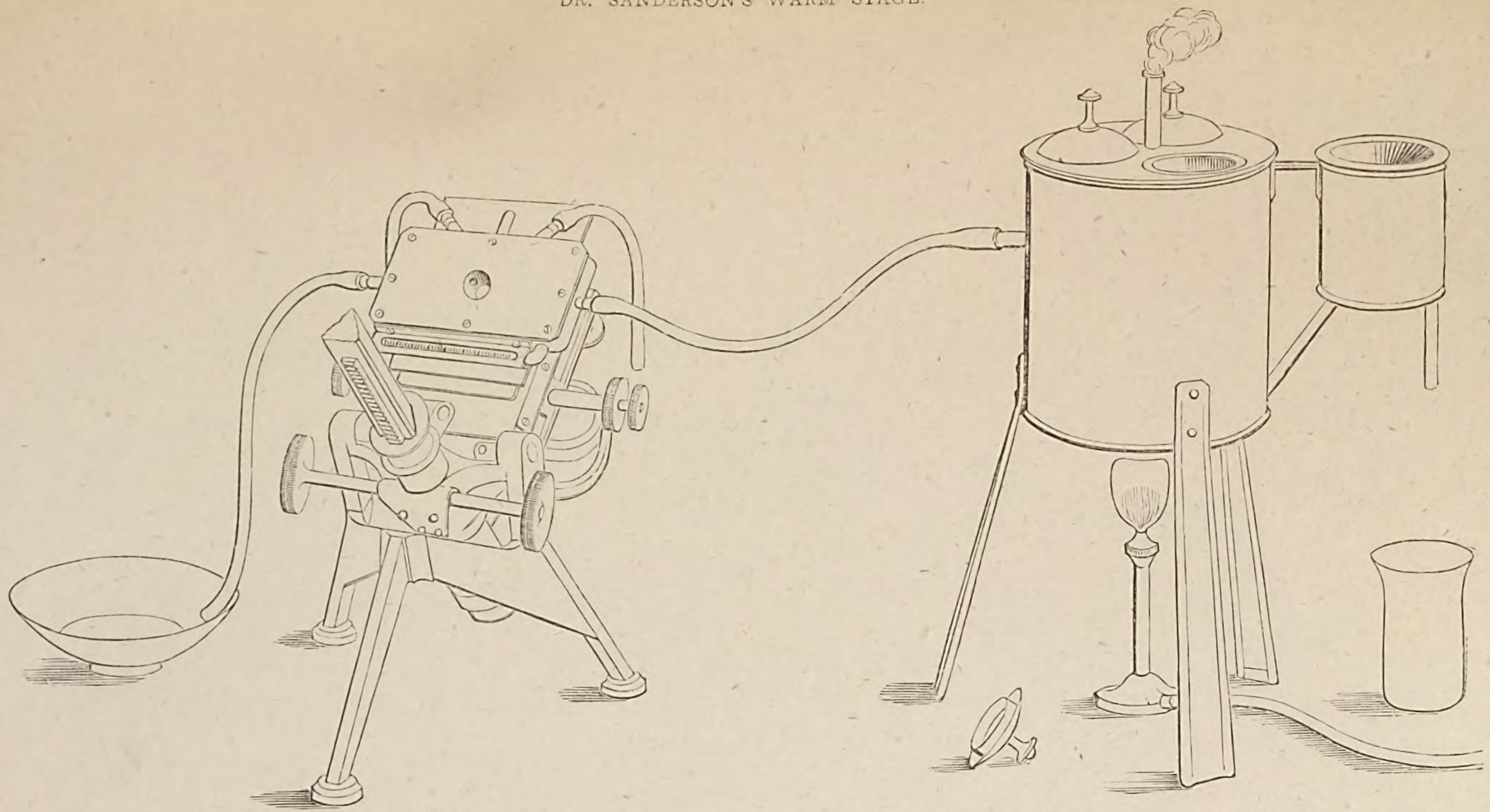
168. Of Detecting Ammonia in the Breath.—Ammonia was first detected in the expired air by the Rev. J. B. Reade, about fifteen years ago, and Dr. Reuling has obtained evidence of the presence of a large quantity in typhus fever, pyemia, and poisoning by urea. In the latter condition it has been detected in the breath, and also in the blood, by Dr. Frerichs, who attributed the coma to the ammonia in the blood, instead of to the accumulation of urea, as had been supposed by previous observers. This subject has been more recently investigated by Dr. Richardson. His researches gained the Astley Cooper prize for 1856.*

Dr. Reuling proposed to test the expired air for ammonia by hæmatoxylin, which forms, with this substance, a rose-red colour; but the best plan is the one originally employed by the Rev. Dr. Reade and recommended by Dr. Richardson. An ordinary microscopic glass slide is moistened on one side with hydrochloric acid and breathed upon. The ammonia combines with the acid, and a chloride of ammonium is formed which crystallizes in crosslets and dendriform masses. Dr. Richardson recommends the following modification of this process :— An instrument of the form of a straight breast-pump is employed to breathe through ; a drop or two of hydrochloric acid is placed in the bulb, and a perfectly clean slip of microscope glass placed across the trumpet-extremity of the tube, and secured by an India-rubber band. The alkali, as it passes over the bulb, combines with the acid, but some

* "The Cause of the Coagulation of the Blood," by B. W. Richardson, M.D. Churchill, 1858.

of the acid and alkaline vapours pass over together and condense on the microscope glass. As this becomes dry, crystals are formed. A very considerable excess of ammonia may be detected in the breath in typhus fever and many other low conditions of the system, but even in health traces are always to be found. Dr. Richardson found but one exception to this, in the case of a gentleman who lived entirely on vegetable food. Ammonia is more abundant in the breath of healthy persons after fatigue than in the morning after sleep; and in hot weather a much larger proportion is expired than in cold weather. Mr. Lens Aldous made some excellent drawings of the microscopical characters of the matters from the breath, condensed on a glass slide, the interest of which was enhanced by the fact that the eyes and hands concerned in the production of the drawings had not lost their power at the age of eighty, though they had been very hardly worked up to that period of life. Crystals of chloride of ammonium from the breath are represented in fig. 6, pl. XXIII, p. 196.

DR. SANDERSON'S WARM STAGE.



Warm stage, with thermometer and arrangement for supplying hot water. The apparatus figured is made by Mr. Hawksley, of Blenheim Street, Bond Street, p. 118.

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CHAPTER IX.

On examining Deposits from Fluids.—Test Tubes.—Pipettes.—Conical Glasses.—Wash Bottle.—Funnels and Filtering.—Cells for Examining Deposits.—On Removing Deposits for Examination.—Of Collecting a very small quantity of Deposit.

On Examining Deposits from Fluids.

The most important pieces of apparatus required in the examination of deposits which subside from fluids, are the following:—Test tubes, pl. XVI, figs. 7, 8, pipettes of different sizes, pl. XVI, fig. 5, conical glasses, pl. XV, figs. 1, 2, wash-bottle, pl. XVI, fig. 6, watch-glasses, funnels for filtering, pl. XVI, fig. 11; and cells in which the deposit may be subjected to microscopical examination, pl. XVI, figs. 9, 10.

169. Test Tubes.—The observer should be provided with several test tubes, varying in length from five or six inches to an inch and a half, or even less. The smaller tubes, besides being adapted for examining small quantities of fluids or deposits, are very convenient for preserving small quantities of deposits immersed in a preservative solution.

In boiling a specimen of urine the test-tube may be held by twisting a piece of paper three or four times folded round the neck, so as to serve for a sort of handle; or a little support made of wire, and mounted in a wooden handle may be used; or the tube may be placed through the smallest ring of the small retort-stand represented in pl. VI, fig. 1, p. 74, and in this manner exposed to the action of the lamp.

170. Pipettes.—The pipettes required in microscopical examination should be of various sizes, according to the depth of the vessel which contains the deposit and the diameter of its orifice. When it is required to remove some of the deposit from the bottom of a bottle with a narrow neck, we shall want a pipette of very small calibre. If the deposit be very thick and viscid, the pipette must have a wide orifice, or it will not enter it. The orifice of the pipettes should vary from the tenth to the eighth of an inch in diameter. Pipettes are made of common glass tube of various sizes. The tubing is to be drawn out slightly in the blowpipe, at the point where the orifice of the pipette is desired, in order to make it a little narrower than the tube itself.*

* Glass tubing adapted for making pipettes, may be purchased of the operative chemists, or of Messrs. Powell, Whitefriars.

When cold a scratch is to be made with a triangular file and the tube broken, the cut end being slightly heated in the flame of the spirit-lamp, to round the sharp edge. The top of the pipette should be slightly bent over in the form of a lip, and made perfectly smooth, so that it may be completely covered with the forefinger, while the middle finger and thumb are placed on either side of the tube immediately below the ring, pl. XV, fig. 1.

It is convenient to have a sort of collar to the pipette, about two inches from the top. This will prevent the finger and thumb from slipping when the instrument is used, pl. XV, fig. 1. Occasionally a pipette, the end of which is slightly bent round, will be found useful, pl. XVI, fig. 5; and sometimes when we wish to decant a considerable quantity of fluid from a watch-glass, &c., a pipette, upon the stem of which a bulb has been blown, will be of service. Various other forms of pipettes have been employed, but the above will be found most useful to the microscopical observer. A small pipette with a narrow opening is convenient for removing any superfluous fluid which may escape outside the thin glass cover when preparations are being mounted in fluids and preserved in cells.

171. Conical Glasses.—Glasses of the shape figured in pl. XV, figs. 1, 2, are the most convenient vessels in which to allow deposits to subside. After the fluid has stood for some time, the deposit will have collected in the narrow portion of the glass, and however small in quantity, it may be very easily removed with the pipette. These glasses can be obtained of various sizes. In choosing them, it is better to select those in which the narrow part terminates in a slightly rounded extremity, for many will be found to terminate in a sharp pointed end, too narrow to force into it even a wire, for the purpose of cleaning, while others have a little prominence in the bottom around which the deposit collects, and in this case it is with difficulty removed by the pipette. If the deposit is allowed to dry in such badly-made glasses, it is hardly possible to clean it. The very useful glass for allowing deposits to collect in, represented in fig. 2, was designed by Dr. Budd, and is of great use in examining urine and other fluids, as the specific gravity may be taken, and the specimen then allowed to stand in the same glass without the trouble of transferring it to another vessel, as is necessary if the ordinary upright jar be employed for taking the specific gravity.

172. Wash-Bottle.—This simple piece of apparatus, which is ordinarily used by chemists, is of great use to the microscopical observer. He will find it very convenient for washing away the parenchymatous part of tissues in order to leave the most fibrous portions, removing epithelium from the surface of membranes, &c. It is employed by the chemist chiefly for washing precipitates on

filters, &c. The wash-bottle is made with an ordinary bottle or glass flask, having a moderately wide mouth. Two tubes bent, as shown in pl. XVI, fig. 6, are accurately fitted into a cork adapted to the neck of the bottle. Upon nearly filling the bottle with water, and blowing through the shorter tube, the fluid will be projected from the capillary orifice of the longer one in the form of a fine jet, which may be directed upon any desired point.

173. Funnels; Filtering.—The funnels required in microscopical examination are very small. Those of about two or three inches in diameter are large enough for most purposes. Glass funnels are the cheapest and the best. The funnel is supported in the small retort-stand, pl. XVI, fig. 11, or upon a tripod. The filtering paper may be obtained already cut in packets of circular pieces of any size required. One of these is folded in the manner shown in pl. XVI, fig. 12, when used. Before filtering, the filter should always be moistened with a few drops of water, or with a fluid of the same nature as that which is to be filtered. A single drop of fluid may be filtered by placing a narrow piece of filtering paper of a ⋖ form close to the drop on a glass slide, and inclining it so that the clear fluid runs downwards from the apex of the ⋖.

Solutions of *crystalloid* substances may be separated from *colloids* by dialysis. For this purpose the mixed fluids are placed in a vessel, the bottom of which is formed of bladder or parchment-paper. The lower surface of the dialyser is in contact with water in a basin. Under these circumstances the crystalline matters pass through and diffuse into the water beneath, while the colloid will be retained in the dialyser.

Straining through muslin is sometimes a convenient method of separating fine and coarse particles from each other, or for separating a crystalline deposit from viscid mucus. By projecting a stream of water from the wash-bottle, the crystals may be washed through the muslin into a vessel placed beneath to catch them, while the mucus remains behind. In the separation of starch particles from gluten, a similar plan may be pursued. The muslin may be tied over a glass or funnel with a piece of thread, or it may be conveniently fixed in its place by one of the vulcanised India-rubber rings commonly sold at stationers' shops.

174. Cells for the Examination of Deposits.—The thin glass cell, pl. XVI, fig. 9, will be found very convenient for the examination of deposits from fluids, especially when they exist only in small amount. If the deposit is very abundant, it will only be necessary to place a small quantity upon a glass slip, and cover it with thin glass. For the examination of urine, sputum, &c., I have been in the habit of using the animalcule cage.

Animalcule Cage.—The advantage of this apparatus consists in the facility with which the depth of the stratum of fluid to be examined may be altered, according to the quantity of the deposit which it contains. This is a point of great practical importance when the amount of sediment is very small, for by submitting only a very thin stratum of fluid to examination, we might often overlook the presence of a small quantity of an important deposit, such as a few fat-cells, or small crystals of oxalate of lime in urine. If, on the other hand, the deposit be very opaque and abundant, the cover may be pressed down so as to come very nearly into contact with the glass upon which it is placed, and an extremely thin stratum may in this manner be examined. The most useful forms of animalcule cages for examining urinary deposits are represented in pl. II, fig. 8, p. 22, pl. XVI, fig. 10. In cases where exceedingly thin objects are to be examined with the aid of very high powers, and it is necessary to prevent the too firm pressure of the thin glass, a fragment of hair, or small pieces of paper or card, according to the thickness of the object to be examined, may be introduced beneath the thin glass.

175. Removal of the Deposit from the Vessel containing it.—This is effected as follows:—The upper end of the pipette is to be firmly closed with the forefinger, while the tube is held by the thumb and middle finger, and the lower end plunged through the fluid to the bottom of the vessel containing the deposit, pl. XV, fig. 1. If the forefinger be now raised very slightly, but not completely removed, a few drops of the fluid with the deposit will rush up into the tube. When a sufficient quantity for examination has entered, the forefinger must again be firmly pressed upon the upper opening, and the pipette carefully removed. A certain quantity of the deposit is allowed to flow from the pipette on to the glass slide or cell, by gently raising the forefinger from the top. The deposit is then covered with the thin glass cover, and subjected to examination in the usual way.

176. Method of Collecting a very small quantity of a Deposit from a Fluid.—When the quantity of deposit is very small, the following plan will be found of practical utility. After allowing the lower part of the fluid which has been standing, to flow into the pipette as above described, and removing it in the usual manner, the finger is applied to the lower opening, in order to prevent the escape of fluid when the upper orifice is opened by the removal of the finger. The upper opening is then carefully closed with a piece of cork. Upon now removing the finger from the lower orifice, the fluid will not run out. A glass slide is placed under the pipette, which is allowed to rest upon it for a short time. It may be suspended with a piece of string, or supported by the little retort-stand. Any traces of deposit will subside to the lower part of the fluid, and must of necessity collect in a small

drop upon the glass slide, which may be removed and examined in the usual way, pl. XV, fig. 3.

Another plan is to place the fluid with the deposit removed by the pipette, in a narrow tube, closed at one end, the bore of which is rather less than a quarter of an inch in diameter. This may be inverted on a glass slide, and kept in this position by a broad elastic India-rubber band. The deposit, with a drop or two of fluid, will fall upon the slide, but the escape of a further quantity is prevented by the nature of the arrangement, pl. XV, fig. 8.

177. Separation of the Deposit from the Fluid in which it was suspended, for Preservation.—After allowing time for the complete subsidence of the deposit, the supernatant fluid is poured off, and the glass filled up with water, or some fluid which corresponds in density to that which was removed, as glycerine, saline solutions, &c., in cases in which the endosmose of water into structure is to be feared. After again allowing time for the subsidence of the deposit, the operation of pouring off the fluid is repeated, and more water, or the preservative solution, added, and again poured off, until the deposit is considered to be free from the original fluid. Two or three washings generally suffice. In this way a deposit may be thoroughly saturated with any fluid in which it is to be preserved.

After being thoroughly washed, the deposit may be removed with a pipette in the usual manner, and placed upon a slide, or in a small test tube, which may then be corked up and labelled. The latter plan is the most satisfactory with which I am acquainted for preserving small quantities of deposits, and if the tube be nearly filled with the preservative fluid, the deposit will keep for any length of time. In this manner I have kept many delicate specimens for twenty years.

Of separating the Fine from the Coarse Particles of a Deposit.—This is readily effected by stirring the whole of the deposit up with some water. When a short time has been allowed for the subsidence of the densest particles, the fluid is poured off into another vessel. After a short period has elapsed, all but the deposit is again poured off into a third and fourth vessel. In this manner several different sediments are obtained, each containing particles of different size and density, which may be subjected to examination or mounted separately.

CHAPTER X.

Of the Chemical and Microscopical Examination of the Solids and Fluids of the Animal Body.—Reaction.—Of taking Specific Gravities.—Evaporation, Incineration, &c.—Dialysis; Colloids, Crystalloids, &c.—Apparatus required for Chemical Investigation.—Microscope for Examining Substances Immersed in Corrosive Liquids.—Method of Examining Objects under the Influence of Heat and Cold.—Reagents.—Method of Applying Tests to Substances intended for Microscopical Examination.—Bulbs and Tubes with Capillary Orifices.—Application of Reagents to Minute Quantities of Matter.—Testing for Carbonates.—Effects of Reagents upon Animal Structures.—Effects of Acids and Alkalies.—Of obtaining Crystalline Substances from the Fluids and Textures of Animal Bodies.—Influence of some Constituents upon Crystallization.—Separating Crystals from Animal Substances.—Examination of Crystals under the Microscope.—Preservation.—Urea.—Creatine.—Uric Acid.—Hippuric Acid.—Lactic Acid and Lactates.—Taurine.—Leucine.—Tyrosine.—Fatty Matters.—Cholesterine.—Myelin.—Excretin.—Crystallizable Matters from the Blood.—Crystallization of Bile.—Of Removing Stains from the Hands.

By the aid of the microscope we are enabled to distinguish many substances with certainty, but amorphous particles are very often met with, the nature of which it is impossible to ascertain by microscopic investigation only. It is therefore necessary to study the effect of certain reagents upon the substances under the microscope. We may learn by microscopical examination that a texture is *granular*, *fibrous*, *opaque*, *perfectly clear*, &c., but nothing of its physical and chemical properties can be ascertained by simply looking at it. Since the same appearances are manifested by several different substances, it is necessary to resort to a chemical examination to determine the nature of many things which come under examination. If, however, the composition of any body having well-defined microscopical characters has been once conclusively determined, we shall be enabled afterwards to recognize it by resorting to microscopical examination. Every specimen of granular matter requires chemical analysis, which may be conducted while it remains upon the

glass slide, and the reactions induced may be studied under the microscope.

In almost every branch of microscopical enquiry, the greatest assistance is derived from the use of chemical reagents. By a knowledge of the behaviour of certain substances with particular chemical reagents, and the application of this information to microscopical investigation, we are often enabled to distinguish peculiarities of structure, to ascertain the chemical composition of minute quantities of matter, and to demonstrate clearly the existence of particular compounds in the animal frame with the greatest certainty, some of which would probably entirely escape our observation, if we subjected them separately to the most careful chemical analysis, or to the most searching microscopical examination.

Some bodies always produce well-recognized crystals when treated with certain chemical reagents, and we know that although there may be in nature other crystals of precisely the same form, but of a different composition, these latter could not have been produced under the circumstances present, and hence in such a case we may sometimes feel as confident of the nature of the substance as if an ultimate analysis of it had been performed.

The application of chemical analysis to microscopical investigation has thrown a new light upon the nature of many physiological changes which are constantly taking place in organized bodies in health, and has enabled us to investigate more satisfactorily the modifications which occur when these processes are subjected to the influence of conditions which counteract healthy actions. Such matters are of the deepest interest to us as practitioners of medicine. In the various forms of disease which are constantly being brought under our notice, we ought to study as minutely as possible the nature and course of morbid actions, which it is our duty to investigate fully. From what we learn by scientific research we may be led to suggest means to modify or counteract morbid actions, and may, perhaps, even be able to prevent their occurrence in other cases.

The laboratory is a very necessary adjunct to the dissecting-room, the museum, the post-mortem room, and the clinical wards of our hospitals; and he who desires to employ all the means at present at our disposal for unravelling the mysteries of disease, in order to form a correct diagnosis, or enable him to recommend the right course of treatment, will do well to make this particular branch of chemistry, with microscopical examination, an essential part of his study.

The works of Vogel, Schmidt, Scherer, Hœfle, Gorup von Besanez, Hoppe Seyler, Kühne, and others, which have been published within the last twenty years, have done much to advance our knowledge of animal chemistry; while those of Golding Bird, Schwann, Robin and Verdeil,

and Lehmann, and the excellent Atlas of plates by Dr. Funke, show the vast importance which the combined methods of chemical and microscopical investigation are very fast assuming.*

It is not within the compass of the present work to do more than refer to the general principles upon which chemical examination is conducted, and to give examples of those processes which are of the greatest importance to the student of medicine, and which he may be called upon to perform in the practice of his profession.

As an instance of the great advantage of the application of a few simple tests to specimens under microscopical investigation, I may refer to the different effects of ether upon fat globules (which are so commonly found in different tissues), and crystalline bodies composed of phosphate or carbonate of lime, which sometimes resemble oil globules so nearly in refractive properties, in form, and in general appearance, as to have led to many mistakes. The application of a drop of ether has no effect whatever upon the latter, but it dissolves the former. If, however, the oil globule is covered with a membrane which prevents the action of ether upon it, it is necessary to add a little acetic acid or a drop of solution of potash or soda, in order to dissolve the membrane, when the ether will at once act upon the fat. This instructive observation may be repeated upon the oil globules in a drop of milk. Phosphate of lime is readily soluble in dilute acids, while fat is not acted upon by these reagents. Not unfrequently organic material is deposited with the phosphate of lime, so that it is necessary to allow the globules to soak for a few minutes in the acid before concluding that it exerts no action upon them. By such simple proceedings we are enabled at once to decide a very important question, and one that has led to some discussion and difference of opinion, in consequence of the solubility or insolubility of the globules in acids and ether not having been clearly proved. The detection of the presence of mere traces of urea, uric acid, and other substances in different tissues and fluids by the appli-

* "Anleitung zum Gebr. des Mikroskopes zur Zooch. Anal. u. zur Mikroskop. Chemisch. Untersuch." Dr. Julius Vogel, 1841. "Chemische und Mikroskopische Untersuchungen zur Pathologie," Dr. J. J. Scherer, Heidelberg, 1843. "Entwurf einer Allg. Untersuchungsmethode der Säfte u. Excrete des Thierischen Organismus," Dr. Carl Schmidt, 1846. "Chemie und Mikroskop am Krankenbette," Dr. Hœfle, 1850. Franz Simon's "Animal Chemistry," translated by Dr. Day, for the Sydenham Society. Becquerel and Rodier's "Pathological Chemistry," translated by Dr. Speer. "Physiological Chemistry," Dr. Lehmann, translated by Dr. Day, Cavendish Society, 1851. "Atlas of Physiological Chemistry," Dr. Otto Funke, Cavendish Society, 1852. "Traité de Chimie Anatomique et Physiologique," Robin et Verdeil. "Urinary Deposits," Dr. Golding Bird, new edition by Dr. Birkett, 1857. Bowman's "Medical Chemistry." "Anleitung zur Zoochemischen Analyse," Gorup-Besanez. "Lehrbuch der Physiologischen Chemie von Dr. W. Kühne." Bloxam's "Chemistry." "Outlines of Physiological Chemistry," by Dr. C. H. Ralfe.

cation of reagents, and subsequent microscopical examination, will be referred to in the present chapter.

The Chemical and Microscopical Examination of Animal Solids and Liquids.

Preliminary Observations.—In the first place we should note carefully the general characters which the substance exhibits; its form, colour, size, weight, hardness, &c.; and the colour, density, fluidity, transparency, tenacity, &c., in the case of liquids. Portions of solid textures, and the deposit from fluids must be subjected to microscopical examination.

178. Reaction.—The reaction of any moist substance is found out by testing it with a piece of blue and reddened litmus paper. If the matter be dry, or the reaction of a vapour is to be tested, the paper must be first moistened with a drop of distilled water.

The *blue paper* is *reddened* by *acids*. If the *acid reaction* is due to the presence of *carbonic acid*, the blue colour will be restored upon gently warming the paper over a lamp upon a glass slide, or upon a warm plate.

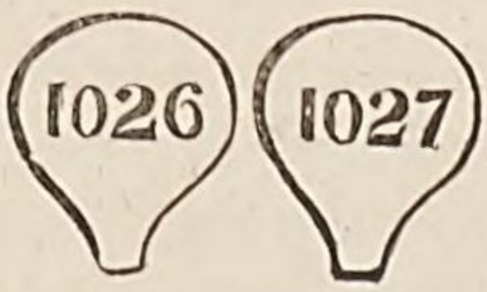
An *alkaline reaction* may depend upon the presence of *volatile* or *fixed alkali*. The *red paper* is turned *blue* by alkalies. Reddened litmus paper is prepared by adding a very small quantity of acetic acid to the infusion of litmus into which it is to be dipped. As the change of turmeric is only visible when the alkaline reaction is very decided, it is not much employed in animal chemistry.

The red colour is restored upon warming the paper which had been rendered blue by the presence of volatile alkali (ammonia or carbonate of ammonia), while it is not restored if the change is produced by the presence of a fixed alkali (potash, soda, or their carbonates, or an alkaline phosphate, &c.).

179. Specific Gravity.—*Solids.*—The specific gravity of animal solids may be taken in two ways. *First.* By weighing in air, and afterwards in water, which is the process usually followed, and that which affords the most accurate result. The precautions necessary to be observed in carrying out this process will be found in "Bowman's Practical Chemistry," and other analytical works on chemistry. *Secondly.* The specific gravity of solids may be obtained by placing small portions in certain saline solutions, the specific gravity of which has been previously ascertained by experiment: this latter method has been employed lately for ascertaining the specific gravity of the brain in different cases of disease.* The solutions are prepared in considerable quantities at a

* Dr. Bucknill "On the Specific Gravity of Cerebral Substance"—(Lancet, 1852). Dr. Sankey in the "British and Foreign Medico-Chirurgical Review," Jan. 1853, page 40. Dr. Aitkin, "Glasgow Medical Journal," No. I, 1853, and "The Science and Practice of Medicine."

time, and kept in large bottles numbered according to the specific gravity of the fluid in each. The strong solution of the salt is first prepared, and this is diluted with such proportion of water as will make several different mixtures, varying in specific gravity from 1030 to 1052. The density of the solutions may be ascertained by the specific gravity bottle, by the urinometer, or by the aid of the little glass bulbs, represented in the figure in the margin. Several glasses are nearly filled with the solutions from different bottles, and arranged in regular order. The piece of tissue is thrown into one, and, if it sinks, it must be placed in the fluid of the next higher specific gravity, and so on, until it neither sinks towards the bottom nor rises to the surface, when the specific gravity marked upon the bottle will correspond to that of the substance itself, since a solid will displace an equal bulk of a solution which is of the same specific gravity as itself. The soluble substances employed for making the solutions, may be sugar, various salts, glycerine, and other materials, which do not exert any chemical action upon the tissue, whose specific gravity we wish to determine. Dr. Aitken recommends sulphate of magnesia, as the action of this salt on the cerebral tissue is very slight.



Bulbs for taking the specific gravity of fluids.

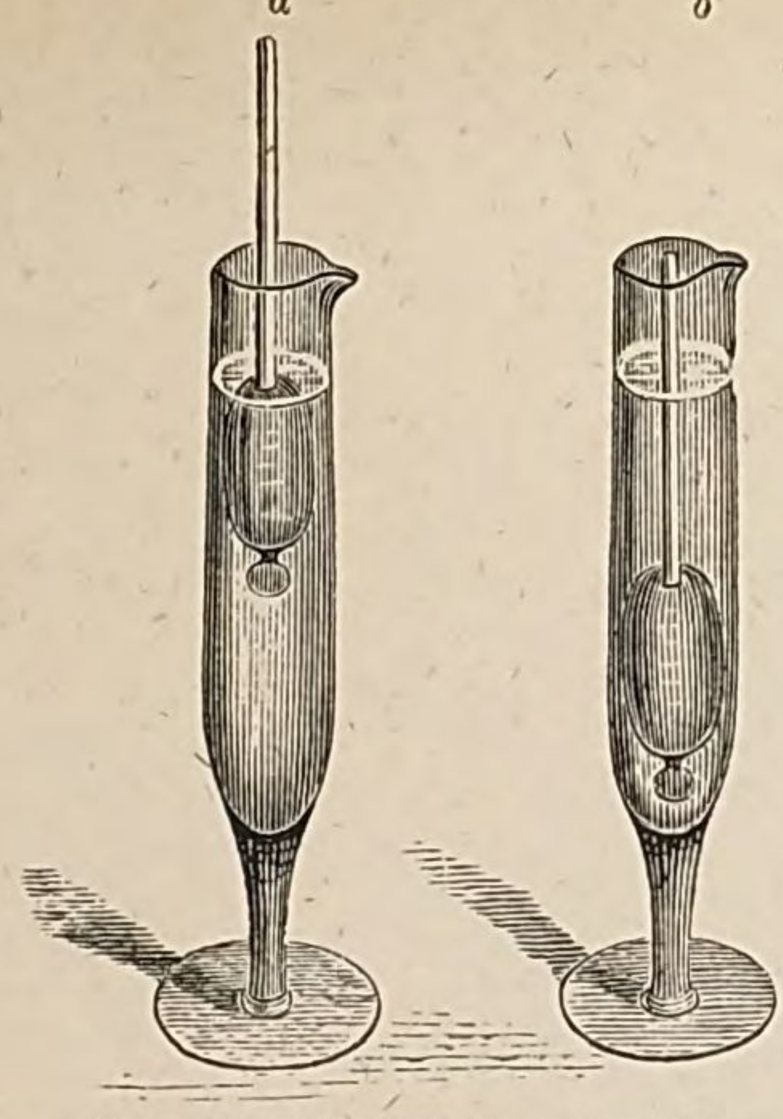
Specific Gravity.—Liquids.—First. By the converse of the last operation, namely, by placing little glass bulbs, the specific gravity of which is marked upon them in the solution, the density of which we wish to know, until one is found which neither sinks nor swims. This will indicate the specific gravity of the fluid. This method is neither so correct, nor so easily applicable to general purposes as the two following.

Secondly, by the *hydrometer* or *urinometer*. The number which is on a line with the surface of the fluid, when the instrument comes to rest, indicates its specific gravity. This method is tolerably correct, if the observer is careful to obtain the best instruments; but many which I have examined, indicated a specific gravity eight or ten degrees from the truth. The correctness of the hydrometer or urinometer should always be verified by the specific gravity bottle. It may be remarked that the degrees marked upon the stem should gradually diminish in length from above, downwards, pl. XV, fig. 7 B. If they are equal as in A, the instrument may at once be pronounced incorrect, without resorting to an experiment. The degrees at the lower part of the stem should be shorter than those near the top, as shown in B. The necessity of this inequality in the degrees will be rendered evident by referring to the figures in plate XV. In fig. 2 *a*, representing a dense fluid, the stem is of course almost entirely above the surface of the liquid, but in *b*, a fluid supposed to be but little heavier than water, only a very small piece of the stem rises above the surface.

Fig. 1.

Conical glass for allowing deposits from fluids to subside, p. 150.

Fig. 2.

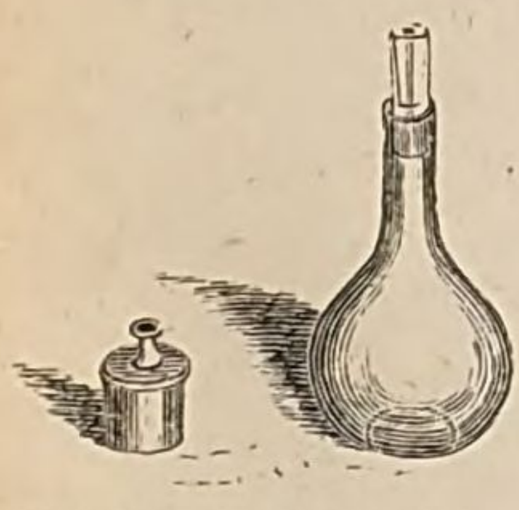


Glasses for collecting deposits and for taking the specific gravity of fluids. pp. 150, 158.

Fig. 3.

Mode of collecting deposit upon a glass slide from fluid contained in a pipette. p. 152.

Fig. 4.

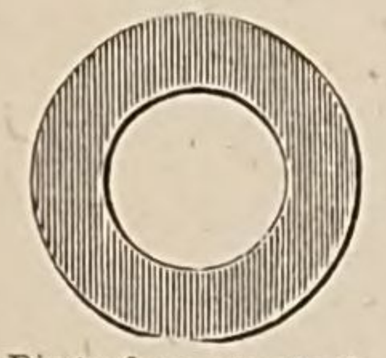


Specific gravity bottle, with tubulated stopper and counterpoise. p. 158.

Fig. 5.

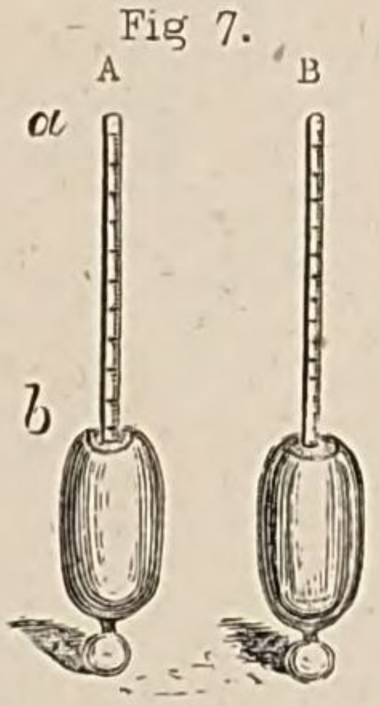
Small water bath, with basin.

Fig. 6.



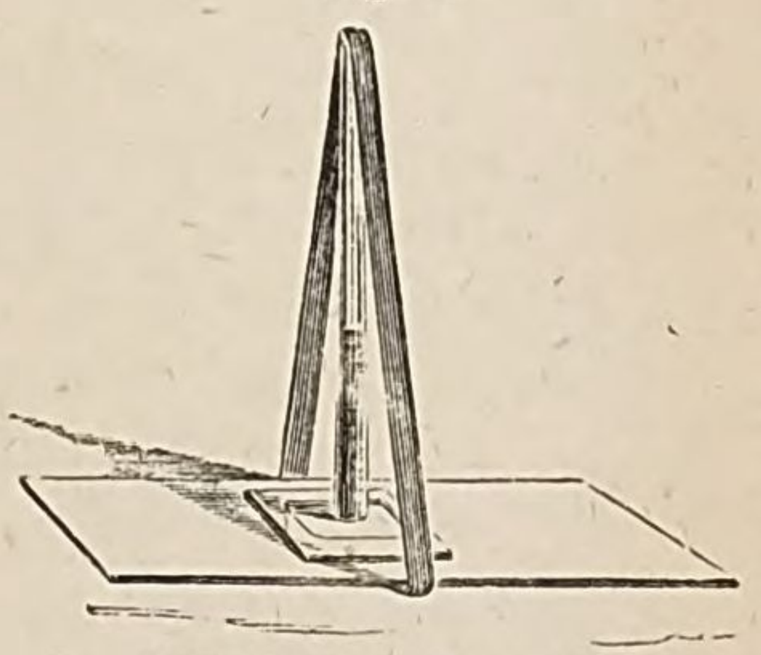
Rings for water bath, for adapting the bath to hold any size basin. p. 159.

Fig. 7.



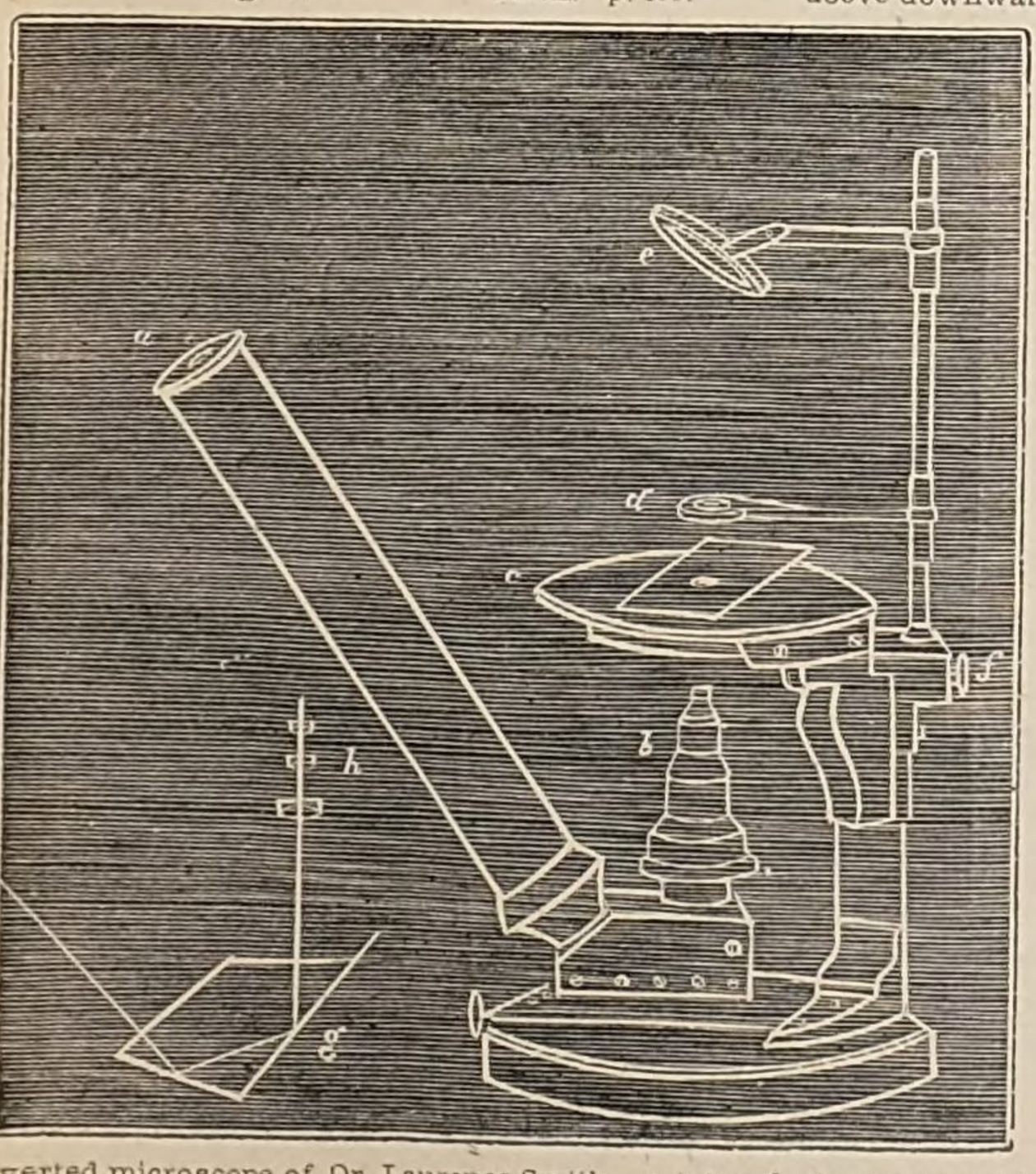
Urinometers. A, an imperfect instrument, in which the degrees are equally divided. B, a good instrument, in which they gradually diminish from above downwards, p. 158.

Fig. 8.



Collecting deposit. p 153.

Fig. 9.



Inverted microscope of Dr. Laurence Smith. *a*, tube of microscope, with eye-piece *b*, object glass on a box, which contains a prism resembling that marked *g*. *c*, stage, with slide upon it. *d*, support on which polarising apparatus or condenser may be placed. *e*, mirror. *f*, screw which elevates or depresses the stage in focussing. *g*, prism, showing the direction in which the rays of light passing through it are refracted. *h*, position of achromatic object glass. p. 162.

Fig. 10.

Apparatus for examining objects while exposed to heat. The chimney through which the heated air passes is made of thin copper without being soldered. Both sides are perforated, the lower one being filled up with a piece of glass, while over the upper opening the slide with the object is placed. p. 162

[To face page 158.

Now, in the first figure, the greater weight of stem above the surface of the fluid, tends to press down the bulb with greater force than the small portion exposed in fig. B. Hence it is necessary that this should be allowed for in graduating the instrument, and the degrees at the lower part of the scale must be shorter than those at its upper part. Mr. Ackland, at Messrs. Horne and Thornthwaite's, Newgate Street, graduates urinometers most accurately.

Thirdly, by the *specific gravity bottle*, which consists of a small glass flask. When quite dry, it is accurately counterpoised in a delicate balance, filled up to a certain point with distilled water, and weighed. The distilled water is then poured out, and it is filled up to the same point exactly with the liquid to be tested, and again weighed. The specific gravity is then readily calculated from these data. Some bottles are made to hold exactly one thousand, five hundred, two hundred and fifty, or one hundred grains of distilled water, and are provided with a perforated stopper, through which the excess of fluid escapes, after the bottle has been filled, care being taken not to include air-bubbles, pl. XV, fig. 4. The outside of the bottle is wiped dry, and the whole weighed. The weight shows the specific gravity at once, upon deducting the weight of the thousand-grain bottle; or, when a five-hundred grain bottle is employed, the amount only requires to be doubled. If the bottle holds two hundred and fifty grains, the weight must be multiplied by four, and so on.

180. Evaporation and Drying.—The evaporation of animal fluids, and the desiccation of animal solids, must always be conducted over a water-bath, or in a little hot water or air oven, otherwise there is great danger of decomposition occurring. For operations upon small quantities, the water-bath represented in pl. XV, fig. 5, will suffice, or the cans of the injecting apparatus used for melting injections made with size may be removed, and basins placed over the holes. A very simple form of water-bath is made by placing a small porcelain basin with a little water in it over the lamp, and upon the first basin, a second containing the substance to be evaporated, pl. VI, fig. 2, page 74.

In endeavouring to obtain crystals of organic substances, it is always advantageous to evaporate the solution over the surface of sulphuric acid under a bell-jar, or, what is better still, in vacuo. And when evaporation has been conducted by heat, it is always desirable, before weighing, to let the vessels cool over sulphuric acid. Extracts may also be kept in this way from day to day without absorbing fresh moisture. In some instances, the evaporation may be conducted by simply exposing the liquid placed in a basin or watch-glass, and covered lightly with paper, to the air; or, where very slow evaporation is necessary, the watch-glass may be covered over with a bell-glass.

When quantitative analysis is to be performed, much greater care must be observed in the process of drying, which must be completed, if not conducted, in vacuo over sulphuric acid. Drying is one of the most important and difficult operations to be performed in physiological chemistry.

181. Incineration.—By incinerating a small portion of any organic substance, upon a piece of platinum foil, or in a platinum or porcelain crucible we are enabled to ascertain whether it contains inorganic salts, or consists entirely of organic matter. In the latter case the substance leaves only a black residue, which burns off entirely after a short time. In order to obtain the inorganic constituents perfectly free from carbon, it is sometimes necessary to keep the mass, for a considerable time, at a dull red heat. The addition of a drop of nitric acid causes the rapid oxidation of the carbon. If the temperature be too high, the process is often much retarded, in consequence of the fusion of some of the salts, as the phosphates and chlorides, and the inclusion of small masses of carbon, which are thus protected from the action of the atmosphere.

The platinum basin or foil may be supported over the lamp upon a piece of wire bent in the form of a triangle, or upon one of the small rings attached to the spirit-lamp, pl. VI, fig. 1. It may be removed from the lamp with the aid of an old pair of forceps.

182. Dialysis.—Colloids and Crystalloids.—By the valuable researches of Professor Graham many very interesting points with reference to the physical constitution of several substances entering into the formation of the organism have been brought to light. He has shown that substances exist in what is termed a *colloid state*, in which condition they will not permeate a porous diaphragm; while, on the other hand, crystalloid substances will readily pass through such a diaphragm when in a state of solution in water, § 173. The fact is one of great practical importance, and has been most successfully employed for the purpose of separating poisonous matters of a crystalloid nature from their solution in the animal colloids (dialysis). The crystalloids readily diffuse themselves through a large quantity of water, while the diffusive tendency of colloids is very low. Professor Graham has shown that certain *mineral* substances exist in a colloid as well as in a crystalloid form. Hydrated silicic acid and soluble alumina are examples. Perhaps the most interesting example in the living organism of an organic body, which may exist in both conditions, is the material of which the red blood corpuscle consists, which sometimes, as in the case of the Guinea-pig, passes from the colloid to the crystalloid condition soon after it has been removed from the circulation and allowed to become stationary.

Crystalline substances may be dissolved out of the various tissues

by distilled water. The weak saline solution may then be concentrated by evaporation at a moderate temperature over a water-bath, or by being placed over sulphuric acid *in vacuo*. The crystalline material may thus be obtained in its characteristic form. In the living organism in health, crystalloid matters pass through the walls of vessels, the outer part of gland cells, &c., while the colloids are retained by the blood or tissues. In certain altered conditions of the fluids, or of the membranes, or of both, colloid, as well as crystalloid matters, may filter through in a dilute state.

Dialysis may be conducted upon a small scale, by means of the little vessels described in page 118. Dialysis has lately been employed by Mr. George Johnson for making albumen compounds with acid. Mr. Johnson has discovered that if white of egg be placed on the dialyser, and a weak solution of hydrochloric acid below, the latter combines with the albumen, forming a true chemical compound, which gelatinises. Mr. Johnson has prepared many different compounds which are new. See his paper, for which the Daniell Scholarship was awarded in 1874, published in the Transactions of the Chemical Society.

Apparatus.

The chemical apparatus which is necessary for chemical analysis as applied to microscopical investigation is very simple, and the greater number of instruments have already been referred to. The following are among the most important pieces of apparatus :—

A few conical glasses of different sizes. Apparatus for taking specific gravities. Test-tubes of various sizes, arranged on a stand, pl. XVI, figs. 7, 8. Spirit-lamps, with various supports, or, where gas is laid on, a gas-lamp. Small porcelain basins, watch-glasses ; a simple water-bath, pl. XV, fig. 5 ; or, if several evaporations are to be conducted at once, the injecting-can may be used. A small platinum capsule, a strip of platinum foil, a blow-pipe, pipettes, and glass stirring rods, with a box of reagents in small bottles, pl. XVI, fig. 13, and test-papers, complete the apparatus. All these may be obtained, separately, or packed in a box of convenient size.

183. Microscope for Examining Substances immersed in Acids and Corrosive Fluids.—In examining in the ordinary microscope, preparations which require to be immersed in strong acid, it is not easy to prevent the fumes from injuring the brass work of the instrument. Considerable inconvenience is also experienced in examining fluids while hot, as the vapour which rises, condenses on the object-glass, and renders the object invisible. These inconveniences are entirely obviated by the ingenious microscope invented some years ago by Dr. Laurence Smith, of the United States. ("American Journal of Science," second series, vol. xiv, 1852). An instrument of the kind referred to is repre-

sented in pl. XV, fig. 9, in which also the form and position of the prism are shown. By this arrangement the object-glass is always kept perfectly clear, while of course the definition is not in any way interfered with. In order to adapt this inverted microscope to drawing the outline of objects with the glass reflector, page 34, it would only be necessary to have the body fixed at a right angle with the axis of the object-glass, and a camera lucida or neutral tint glass reflector adapted to it.

184. Arrangements for applying Heat to Objects under Microscopical Observation.—By placing a brass plate upon the stage of the instrument just described, and allowing one end to project over the edge so that it may be conveniently heated by a spirit-lamp, any substance may be kept warm upon a glass-slide, while being subjected to microscopical examination. A very simple apparatus, made by me many years ago, is represented in fig. 10, pl. XV. Heated air was allowed to pass through the tube in which a thermometer could be inserted.

Max Schultze has contrived an arrangement, consisting of a brass plate which is fixed by clamps to the stage of the microscope, and extended at the sides so as to form two projecting arms beneath each of which a small spirit-lamp may be placed. A hole was made for the transmission of the light, and close to the place where the slide with the object is situated, is the bulb of a little thermometer, the stem of which is so arranged that the degrees can be easily read off. Dr. Ransom, of Nottingham, has been long engaged in investigations which require the application of heat and cold to the object while under observation. He says—"The mode of using heat for those examinations I have found best so far, is that recommended by Max Schultze, only in order to employ with it cold also, I have ordered one to be made of copper instead of brass, as the former metal is so much better a conductor, and I trust I shall be able with this new stage to preserve an object at any required temperature, and to read off easily the actual temperature which the object has from 30° F. to 160° F."

The best form of warm stage, however, is that of Stricker, which has been improved by Dr. Burdon Sanderson. See § 139, and plate XIV.

Reagents.

The reagents required for chemico-microscopical examination are not very numerous. They should be perfectly pure. Of the greater number only a very small quantity is required; but of alcohol, ether, and one or two other reagents, it is necessary to have half a pint or more.

The usual reagents should be kept in stoppered bottles of about the capacity of two ounces.

185. Alcohol.—Alcohol of different strengths will be required for the purpose of dissolving certain substances, and for separating them from

other constituents, which are insoluble in this reagent. Alcohol when required weak should always be diluted with distilled water, and it is better to prepare a considerable quantity at a time. It is convenient to have two or three bottles which will hold about two quarts each. The strength of each should be written upon a label attached to the bottle.

The importance of alcohol, as a preservative solution, is well known. Within the last few years, the Government has permitted the use of *Methylated alcohol*, which pays no duty. It may only be used for various purposes in the arts, chemical processes, &c. It answers admirably for preserving anatomical preparations, and is a great boon to all engaged in putting up large specimens. Any person wishing to use this alcohol in large quantity, must in the first instance make application to the Board of Inland Revenue, Somerset House, for permission. The application must be accompanied with the names of two respectable householders, who are willing to serve as bond that the applicant only uses the spirit for the purposes stated in his application. The probable quantity required annually must also be stated. It may be obtained at the price of 5*s.* 6*d.* per gallon, sixty degrees over proof, of varnish makers and some chemists.

186. Ether.—An ounce or two of ether will be quite sufficient for microscopical purposes. It should be kept in a stoppered bottle, provided with a glass cap, to prevent loss by evaporation. A little should also be kept in one of the small glass bottles with capillary orifices, § 200, for the convenience of applying it to granules, highly refracting globules, &c., under the microscope. Methylated ether may be used with advantage. *Chloroform* and *benzine* are also required by the microscopist.

187. Nitric Acid should be kept of two different degrees of concentration: one the strongest that can be procured, and another containing about twenty per cent. of the strong acid. This last is the acid most used by the microscopist, especially in separating muscular fibre cells. It is prepared by mixing one part of the strong commercial acid with four parts of distilled water.

188. Sulphuric Acid is sometimes required undiluted, but a small bottle of diluted acid (one of acid to five of water) should also be at hand. The pure colourless acid should always be procured;—it is to be purchased for about 1*s.* 6*d.* a pound, but only very small quantities are required.

189. Hydrochloric Acid may be obtained perfectly colourless. It may be kept in the pure state and diluted as required.

190. Acetic and other Acids.—Two specimens of acetic acid will be found convenient. One, a solution of the strongest acid which can be procured; the other containing about twenty per cent. This is prepared by dissolving one part of the strongest liquid acid, or of the

pure *glacial acetic acid* in four of water. The glacial acetic acid is now commonly employed for photographic purposes, and can therefore be very readily obtained.

Citric, Oxalic, and Lactic Acids have also been recommended for microscopical enquiries. One part of the acid to from 10 to 20 parts of water, makes a solution of a convenient strength.

191. Chromic Acid is usually required very dilute. For the purpose of hardening tissues, a watery solution of a straw colour (from $\frac{1}{4}$ to 2 per cent.) will be required. It is easily prepared by dissolving a little of the crystallized chromic acid in distilled water. It may be kept in solution containing 10 per cent. and this may be diluted as required.

The crystallized acid may be prepared by decomposing 100 measures of a saturated solution of bichromate of potassa, by the addition of 120 to 150 measures of pure concentrated sulphuric acid. As the mixture becomes cool, crystals of chromic acid are deposited, which should be dried and well pressed on a porous tile, by which means the greater part of the sulphuric acid is removed, and the crystals obtained nearly pure.

192. Solution of Potash should be kept of two or three different degrees of strength. One, the strongest which can be obtained; another made by mixing one part of the strong acid with three or four of water. A solution consisting of one part of liquor potassæ to eight or ten of water, will be found of a useful strength for the examination of many preparations.

193. Solution of Soda is generally required very dilute. It may be made by mixing one part of the strong solution of the shops with five or six of water, or, about twenty-five grains of the fused soda may be dissolved in an ounce of distilled water. These solutions, for many purposes, will require to be still further diluted. Lime water and baryta water have also been employed in microscopical enquires.

194. Ammonia.—Solution of ammonia, made by mixing one part of the strongest liquor ammoniæ with three of water, will be found sufficiently strong for all the purposes for which this reagent will be required. The microscopist should always keep a bottle of the strongest liquor ammoniæ.

195. Nitrate of Barytes.—A cold saturated solution of the salt forms a test solution of convenient strength. It should be filtered before use. A solution of nitrate of barytes is employed as a test for sulphuric and phosphoric acids, either free or in combination, as sulphates and phosphates. The precipitated sulphate of baryta is insoluble both in acids and alkalies; while the phosphate of baryta is readily soluble in acids, but insoluble in ammonia.

196. Nitrate of Silver.—A solution of nitrate of silver is prepared by dissolving one hundred and twenty grains of the crystallized nitrate in two ounces of distilled water, and filtering if necessary. Nitrate of

silver is employed as a test for chlorides and phosphates. The *white* precipitate of chloride of silver is soluble in ammonia, but insoluble in nitric acid. The *yellow* precipitate of tribasic phosphate of silver is soluble in excess of ammonia, as well as in excess of nitric acid. For colouring tissues a very weak solution is required. See page 68.

197. Oxalate of Ammonia and other Salts.—Some crystals may be dissolved in distilled water, and, after allowing time for the solution to become saturated, it may be filtered. Oxalate of ammonia is used as a test for salts of lime. Oxalate of lime is insoluble in alkalies and in acetic aced, but soluble in the strong mineral acids. In testing an insoluble deposit for lime, it may be dissolved in nitric acid and excess of ammonia added ; the flocculent precipitate is readily dissolved by excess of acetic acid, and to this solution the oxalate of ammonia may be added. The precipitation of oxalate of lime is favoured by the application of heat. Many deposits of earthy phosphates are dissolved with great difficulty by acetic acid, hence the necessity of first adding nitric acid, as above directed.

Various salts have been employed in examining and preserving animal tissues and morbid growths. Common salt, chloride of calcium, chloride of potassium, alum salts, some of the alkaline sulphates and phosphates, acetates of soda and potash. Arseniates and many metallic salts have been recommended, but I have not, myself, gained any advantage by their use, and have found that where tissues are to be preserved permanently it is better to remove from them the last traces of saline matters. For if these are allowed to remain, decomposition often occurs, and the preparation is spoiled by the precipitation of granules of some slightly soluble salts. Soluble saline matters will always pass out of textures, by diffusion, if the sections be permitted to remain in a comparatively large quantity of distilled water, or weak glycerine, for some time.

The use of osmic acid has been already referred to in page 70.

198. Iodine Solutions.—An aqueous solution is easily prepared by dissolving a few grains of iodine in some distilled water, until the solution acquires a brownish-yellow colour. This will colour ordinary starch grains, and baked or boiled starch of a deep violet colour. Starch grains thus coloured look perfectly black by transmitted light.

An aqueous solution of iodine is sometimes useful for colouring certain substances, cell walls, fibres, basement membrane, &c. Iodine solutions are valuable tests for starch and allied substances. Iodine and strong sulphuric acid are used for detecting *cellulose* and *amyloid* bodies. The sulphuric acid should be added very cautiously, and a weaker solution sometimes acts if the tissue be soaked for a short time.

A stronger solution of iodine may be obtained by employing a solution of iodide of potassium to dissolve the iodine (one grain of iodine and three grains of iodide of potassium, to one ounce of distilled water).

In this way the 'iodine paint' so much employed by medical practitioners is prepared. For testing bodies suspected to consist of starch, the following solution is recommended by Professor Schultze. Zinc is dissolved in hydrochloric acid; the solution is permitted to evaporate in contact with metallic zinc until it attains the thickness of a syrup; and the syrup is then saturated with iodide of potassium. The iodine is next added, and the solution, if necessary, is diluted with water. Professor Busk gives the following directions for preparing this solution: one ounce of fused chloride of zinc is to be dissolved in about half an ounce of water, and to the solution (which amounts to about an ounce fluid measure), three grains of iodine dissolved by the aid of six grains of iodide of potassium in the smallest possible quantity of water, are to be added. (On "Starch Granules."—Transactions of the Microscopical Society, new series, vol i, p. 67). I have employed a solution prepared in this manner, and can speak very highly of its utility. In making it, it is necessary not to *fuse* the chloride of zinc much, or to use a very high temperature, as decomposition is very apt to take place. In testing starch with this solution, it is advisable to add a very little water, as the solution frequently will not act in its concentrated form.

The iodine solutions above recommended may be made with glycerine when required for testing specimens prepared according to the plan I have particularly recommended. The action occurs more slowly, but is very satisfactory, as the gradual change of colour which ensues is characteristic. The material to be tested must be soaked in glycerine before the iodised glycerine solution is added.

Iodised Serum.—Serum of blood has been strongly recommended by Max Schultze for microscopical investigation. It may be kept for months if a piece of camphor be placed in it. The pure serum effused in some cases of ascites may also be employed. About six drops of tincture of iodine or of solution of iodine in hydriodic acid may be added to an ounce of liquor amnii from an embryo calf. Frey recommends a fluid composed of 1 ounce of ovalbumen, 9 ounces of water, 40 grains of chloride of sodium, and 60 drops of the iodine solution. In using these solutions the thinnest sections of tissues that can be obtained should be allowed to soak for some time in the iodised serum. Iodine solutions have been used for tinting, and so rendering distinct very delicate nerve fibres and certain markings in nerve cells.

Method of Applying Tests to Substances intended for Microscopical Observation.

199. Tests kept in Glass Bottles.—The matter to be tested may be placed upon a glass slide, and, if necessary, a drop of water added, to moisten or dissolve it, as the case may be. Only a small drop of the test-solution is required, and it will be found convenient, in applying it

to the object, to take a drop from the bottle by dipping a stirring-rod into it, and withdrawing it immediately. Enough will adhere to the stirring-rod for the purpose required. The rod should not be dipped in the test fluid a second time, without being first well washed in water. If this direction be not scrupulously attended to, there will be great danger of conveying some of the substances intended for examination into the test bottle, in which case the whole contents would be spoiled. Carelessness on this head has led to great inconvenience, and most serious errors have resulted. Objects supposed to exist in the substance examined having been really introduced in the test solution. The observer must always avoid the chance of removing a portion of a deposit on one glass slide, and mixing it with that on another Claws of echinococci, and other minute bodies, in themselves highly characteristic, may be transported, and find their way into deposits in which we should not expect their presence. From such an accident the existence of hydatids might be very erroneously inferred in a case in which no such disease existed, and thus a serious blunder made in diagnosis. Accidents of this kind can always be avoided if ordinary precautions be taken.

In applying a reagent to the matter to be tested the drop should not be allowed to touch the deposit until the rod has been removed. This can be effected by placing the drop near the substance intended for examination, and then allowing it to come in contact with it, either by inclining the glass slide, or by leading it with a thin glass rod, to the matter to be tested.

Without the greatest attention to cleanliness, the microscopical observer will be constantly led into error, and thereby bring discredit upon himself and upon science. Nothing was more common than to find a specimen which we were examining under the microscope covered with a number of starch granules, which had been introduced from without. These were derived from the squares of thin glass which used to be kept in a little starch powder to prevent fracture. An intimate friend showed me one day some microscopic preparations which contained curious bodies of the nature and origin of which he was not aware. Upon examining the slide, I found a number of scales from the wing of a moth, which had no doubt been floating about in the air and had fallen upon the preparations. In all cases, specimens which are about to be mounted should be carefully protected by glass shades, pl. VIII, fig. 1.

200. Tests kept in Glass Bulbs with Capillary Orifices.—By far the most convenient method of applying chemical reagents to minute quantities of matter, is by the aid of a drop bottle. A very minute quantity of the test solution is allowed to issue from a small glass vessel, having a capillary orifice. In this way a quantity much less than a single drop

can be readily obtained, while there is no danger of any portion of the preparation or any impurity being introduced into the test solution.

In order to prepare a convenient vessel for containing the test solution, I blew a small bulb, about an inch in diameter, at one end of a piece of glass tube, and drew out the other in the blowpipe flame to a moderately fine capillary point. A small cap, made either of glass or gutta-percha, was adapted to the open end, pl. XVI, fig. 1. These bulbs were easily filled, by expanding the air within them, by the heat of a spirit-lamp, and then inverting them so that the orifice dipped below the surface of the solution which was to be introduced, and which was already placed in a small capsule. As the bulb cooled, the liquid rushed into it, to supply the place of the previously expanded air. A small bubble of air should, however, be retained in the bulb, by the expansion of which, by the heat of the hand, some of the fluid will be expelled when the bulb is inverted, and the capillary orifice of the bottle placed near the matter to be tested. The bulbs containing the strong acids and alkalies ought to be furnished with glass caps, but gutta-percha will answer for the other tests.

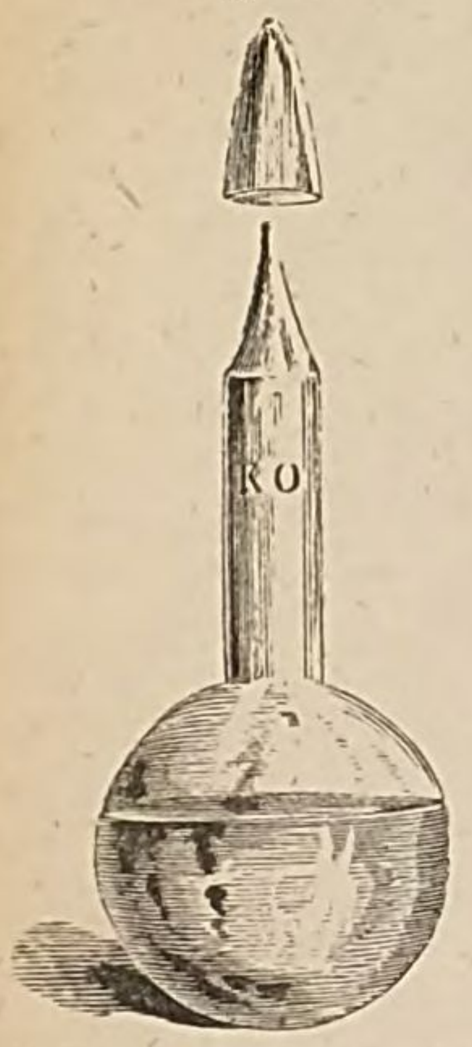
Mr. Highley has had some small bottles made of the form shown in figs. 2 and 3, pl. XVI. These are capped with glass, and as the bottom is flat, they stand very well. They are fitted up in small cases, pl. XVI fig. 13, and will be found exceedingly convenient to the microscopical observer. It is better to have the cap made of a conical shape, corresponding to that of the end of the bottle, otherwise a little of the fluid is liable to collect between the cap and the neck, and may run down the side of the bottle when the cap is removed. Twelve bulbs will be quite sufficient for all ordinary purposes. For the examination of the urine, not more than six or seven will be necessary.

201. Capillary Tubes with India-rubber tied over the Top.—Dr. L. Smith recommends that the tests should be kept in bottles of two ounce capacity, and instead of a stopper, a tube in the form of a pipette, should be made, the upper open end of which was covered with a piece of vulcanised india-rubber, pl. XVI, fig. 4 at *a*. By pressing the elastic membrane while the lower end of the tube was beneath the fluid, a portion of the air was of course driven out, but a little fluid rushed up to supply its place as soon as the pressure was removed. The tube being removed from the bottle, and the india-rubber being again pressed with the finger, a drop, or a portion of a drop, was readily expelled.

Pipettes may also be fitted to perforated corks and used in the same way, the upper end of the pipette being closed with a small cork or an india-rubber bulb adapted to it. Upon the whole I much prefer the dropping bottle described in the last section.

202. Application of the Reagent to Minute Quantities of Matter.—With the aid of the bulbs or other arrangements just referred to, the

Fig. 1.

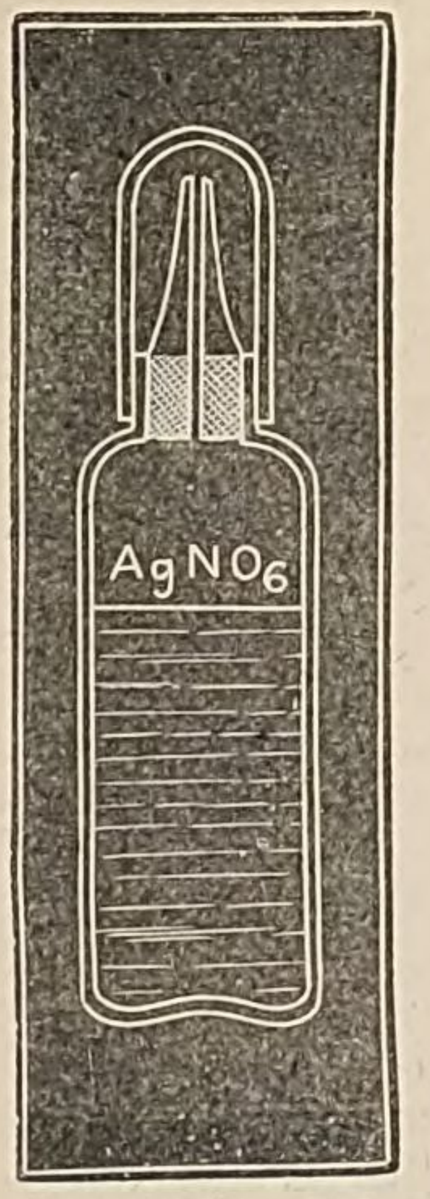


Bulb, with capillary orifice, for testing small quantities of matter. p 168.

Fig. 2.

Small test bottle, with capillary orifice. p. 168

Fig. 3.

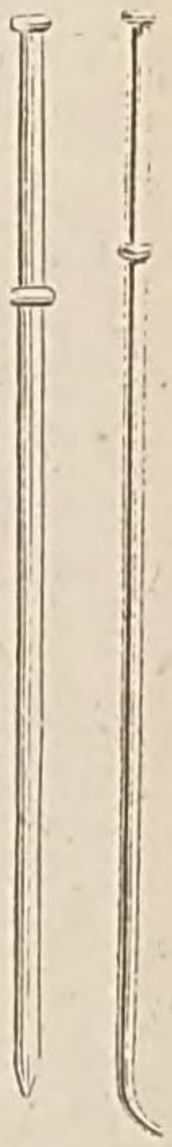


Test bottle, with capillary orifice. p. 168.

Fig. 4.

Pipette, serving as the stopper to the bottle. *a*, vulcanised india-rubber, by pressing which fluid may be expelled from the tube. *b*, ground to fit the neck of the bottle. *c*, the orifice. p. 168.

Fig. 5.

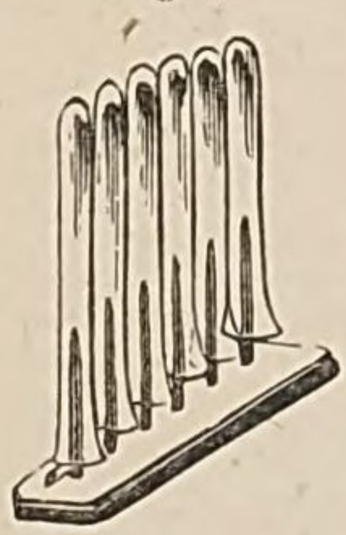


Pipettes p 149

Fig. 6.

Wash bottle, for washing preparations. A stream of water is projected from the orifice of the pipe, when air is blown into the upper tube. § 160.

Fig. 7.



Test tubes on rack § 169.

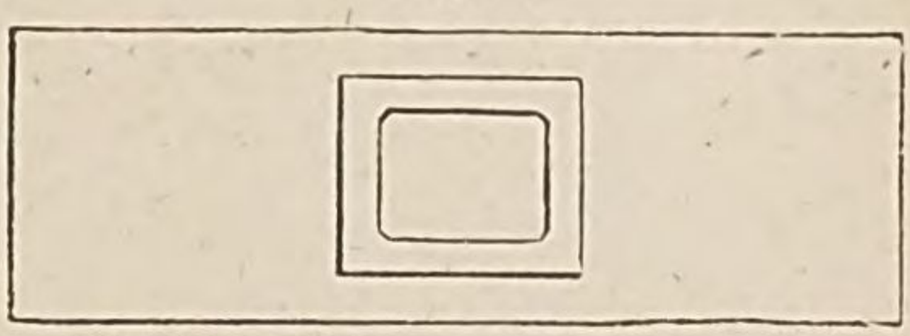
Fig. 7.*

Corked tubes for containing prepared tissues, &c. p. 97.

Fig. 8.

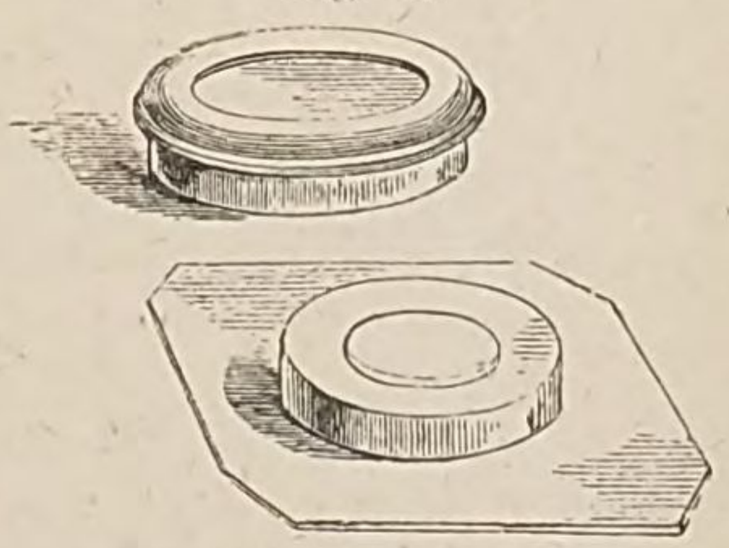
Drainer, for draining test tubes. p 149.

Fig. 9.



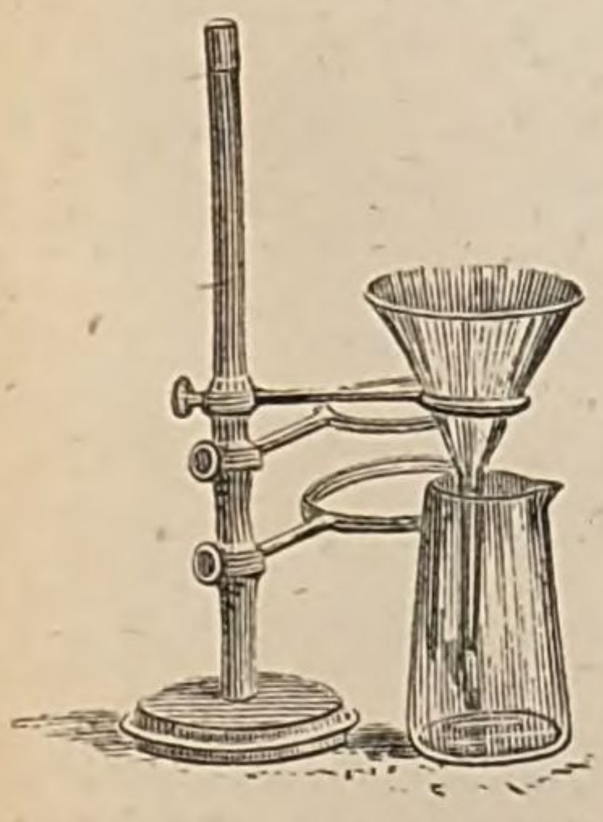
Thin glass cell for examining deposits from fluids, mounting preparations in fluids, &c. p. 151.

Fig. 10.



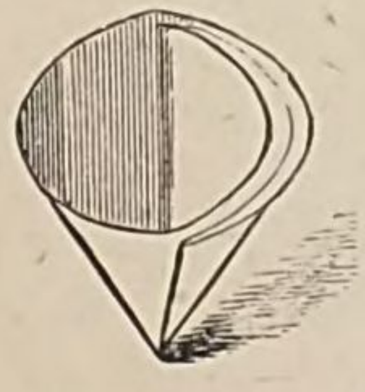
Animalcule cage for examining deposits from fluids. p. 152.

Fig. 11.



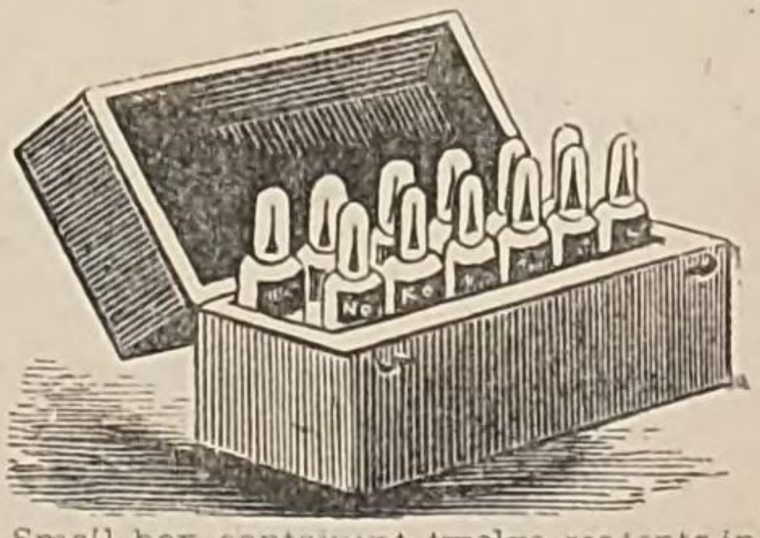
Retort stand, funnel, and glass, arranged for filtering. p. 151.

Fig. 12.



This figure shows the manner in which blotting paper is to be cut and folded for the purpose of filtering. p. 151.

Fig. 13.



Small box containing twelve reagents in tubes, with capillary orifices for testing small quantities of matter. p. 161.

[To face page 163.

most minute traces of different substances may be readily detected. The solution of the substance, consisting perhaps of only one drop, is placed upon a glass slide. This drop may be very easily divided into four or five smaller drops, if necessary, to each of which a separate test may be applied. For instance, suppose we have to examine a minute quantity of the ash of an animal tissue, or of the solid residue of an animal fluid, and we wish to ascertain if it contains carbonates, sulphates, chlorides, and phosphates, and whether phosphate of lime and magnesia are present,—we may proceed as follows:—the portion of ash, which may, perhaps, be half the size of a pin's head, or even less, is removed from the platinum foil, upon which it has been ignited in order to remove organic matter, and placed upon a glass slide. It is moistened with a small quantity of water, and then treated with a minute drop of nitric acid. If effervescence takes place, a carbonate is present. The *acid* solution is then divided into three portions, with the aid of a small stirring-rod, and the solutions, tested as follows:—

1st portion.—If a drop of solution of nitrate of silver gives a cloudy precipitate, *chlorides* are present.

2nd portion.—If nitrate of barytes produces a white precipitate in the acid solution, *sulphates* are present. Upon the addition of excess of ammonia, the precipitate produced by nitrate of barytes will be increased, if *phosphates* exist in the solution. The precipitate of phosphate of baryta is flocculent, and readily distinguishable from that of sulphate of baryta (which is dense and granular), by its solubility in acids.

3rd portion.—If *lime* or *magnesia* be present, in the form of phosphate, a precipitate will be produced upon adding excess of ammonia to the nitric acid solution. The mixture may be stirred a little, with a piece of glass rod or platinum wire, and then allowed to stand for some time. The thin glass cover is now applied, and the precipitate subjected to microscopical examination. *Phosphate of lime* occurs as a granular amorphous sediment, while the *Ammoniaco-magnesian*, or *Triple Phosphate*, is easily found crystallized in a beautiful stellar form, or as minute prismatic crystals.—See "Kidney Diseases, Urinary Deposits, and Calculous Disorders."

203. Testing for Carbonates.—As carbonates are often present in very minute quantity in the ash of organic substances, a slight modification of the plan above indicated may be pursued, and the smallest traces detected. If only a few bubbles of carbonic acid are given off upon the application of the acid to the substance, or if, in consequence of the solubility of the carbonate present, they are evolved very rapidly, they frequently elude observation. In testing for minute traces of carbonates, then, we may proceed as follows:—The portion of ash, deposit, or tissue (as the case may be) is placed upon a glass slide, and lightly

covered with a piece of thin glass. A minute drop of nitric or acetic acid, not too strong, is then allowed to escape from one of the bulbs. This is drawn by capillary attraction between the glasses, and soon comes into contact with the substance to be tested. Any bubbles which may be given off are thus confined, and they may generally be seen clearly enough. In some instances, however, advantage is derived from subjecting the specimen to microscopical examination while the evolution of gas is proceeding. The bubbles set free cannot possibly be mistaken for air-bubbles, which had been included in the interstices of the tissue previously, and afterwards expelled upon the addition of the fluid, because they may be seen to increase gradually in size and number, while the action of the acid proceeds. In testing for carbonates, the possibility of this occurrence, however, must always be borne in mind, and the fallacy carefully guarded against.

Sometimes in testing a deposit of carbonates, the effervescence which is produced upon the addition of the acid, depends upon a little carbonate of ammonia being dissolved in the fluid. We must be careful to ascertain, in the first instance, if the fluid be free from a soluble carbonate, in which case we may conclude the effervescence is caused by the action of the acid on an insoluble carbonate.

Effects of Reagents upon Animal Structures.

The effects of the application of cold strong acids to animal textures are very variable; in some instances the tissue is completely destroyed, while in others scarcely any effect seems to be produced. The mineral acids, except when very dilute, coagulate albuminous tissues, and render their microscopical characters confused and indistinct. Tribasic phosphoric acid, however, is an exception to this statement. Acetic acid also dissolves many substances allied to albumen.

The appearance of some structures is scarcely altered by the application of a strong acid; for instance, the blood corpuscles shrink a little, but retain their circular outline and general appearance for some time after the addition of strong nitric acid. The cells of the epidermis and nail, although turned of a yellow colour, are not destroyed; the latter are separated somewhat from one another, but their outline is often made beautifully distinct. Most of the mineral constituents of the body, insoluble in water, are directly dissolved by the acids.

204. Acetic Acid.—Acetic acid is one of the most useful reagents to the microscopical observer. The glacial acid should be obtained. Acetic acid has the property of dissolving granular matter composed of an albuminous material, and causing the formed material or cell wall to become very transparent. The nucleus usually becomes darker and more distinct. In many instances the action of the acid upon the formed material is peculiar. The entire elementary part becomes much

larger, and its outer part swells up and becomes more pulpy, approaching more nearly in density and refracting power to the solution in which it is immersed. In numerous instances, by adding a saline solution to cells which have been previously rendered transparent by acetic acid, they again contract, and the outline becomes distinct. In some cases, however, the outer part of the cell, or elementary part, is actually dissolved by the acid, and its contents set free. Acetic acid will be required of various strengths, the most useful proportion being one part of the glacial acid to three or five of water. Acetic acid is very frequently used to make epithelial structures transparent, in order that the arrangement of the minute vessels and nerves in papillæ, &c., may be demonstrated, as in the case of the tongue, skin, &c. A slow action is better than a rapid one, and great advantage results from examining the specimens immersed in glycerine.

Sections of preparations which have been hardened by maceration in alcohol, may require boiling slightly in acetic acid before they can be rendered sufficiently transparent. The action of acetic acid on white fibrous tissue is very characteristic, as it converts it into a transparent jelly-like mass, in which a few bioplasts are visible. Upon the yellow element, on the other hand, this reagent exerts no action whatever. The action of acetic acid upon epithelial cells and pus-globules will be discussed in a subsequent chapter.

The action of acetic acid upon any particular tissue, upon any form of cells, fibres, &c., that are subjected to examination, should always be specially noted. Many tissues are quite insoluble in acetic acid, though they are not rendered opaque by it.

Acetic acid may also be employed for testing crystalline bodies, such as phosphates and carbonates. By it we may distinguish phosphate or carbonate of lime from oxalate of lime, all these salts being insoluble in water. Acetic acid dissolves the two former, while it does not affect the latter, even if it be boiled in it.

In my researches upon the arrangement of nerves in various textures, and upon the structure of nerve centres, I have employed a mixture of glacial acetic acid and strong glycerine. I find that in the proportion of ten drops of the acid to an ounce of glycerine, the required action slowly takes place. Specimens may be placed for many weeks or months in a still weaker solution. Gradually the connective tissue acquires transparency, while the finest nerve fibres are rendered slightly granular, in consequence of the oleo-albuminous material being decomposed, and the fatty matter set free as granules or minute globules. The fine dark-bordered nerve fibres retain their smooth appearance, and are scarcely altered by acetic acid in glycerine. If the nerve fibres are very soft it is desirable to add a drop of the glycerine solution of chromic acid to the acid glycerine (p. 103).

Citric, oxalic, and other acids have been recommended for special investigations, but I have not satisfied myself that they possess decided advantages.

205. Dilute Nitric Acid used to be much employed in microscopical research.—An acid composed of one part of acid to two or three of water, forms a good solution for hardening some structures, previous to cutting thin sections. The thin sections, made granular by the acid, may be rendered transparent by being treated afterwards with dilute caustic soda. For demonstrating muscular fibre-cells, nitric acid is a valuable reagent. For this purpose the solution should contain about twenty per cent. of strong acid, and the muscular fibre should be allowed to macerate in it for some days, when small pieces may be removed with scissors, and after being carefully torn up with fine needles, subjected to examination.

When we wish to obtain portions of glandular structures isolated from one another, it is a good plan to soak the tissue for some days in dilute nitric acid (one part of acid to six or seven of water), when the areolar tissue will have become softened, and at the same time the gland structure will be more firm, and may be isolated very readily with the aid of needles. In this manner the gastric glands, the secreting follicles of the pancreas and salivary glands may often be very satisfactorily demonstrated. The so-called fibre-cells of organic muscles are to be isolated in the same way. For this purpose Reichert and Paulsen recommend a 20 per cent. solution.

By boiling animal and vegetable tissues in strong nitric acid, they become destroyed, while any siliceous constituents remain behind unaltered. In this manner, the siliceous skeletons of the *Diatomaceæ* may be separated from the organic matter with which they are combined, and from calcareous and other salts. Nitric acid is also valuable in investigations upon any hard vegetable tissues which require to be softened.

206. Sulphuric Acid.—Hydrochloric Acid.—The pure acids only should be used for microscopical investigation. They may be obtained at most of the operative chemists. Concentrated sulphuric acid causes epidermic structures to swell up very much, and the cells to separate from one another, so that they may be readily isolated. Boiling acid completely dissolves them. In the examination of hair it will be found that, by the moderate action of strong sulphuric acid, the outline of the cells is made very distinct.

Connective tissue becomes converted into jelly, and is dissolved by weak sulphuric acid (one part to 1,000) if kept in it for twenty-four hours, at a temperature a little below 100°. This process has been recommended for isolating the muscular fibres.

Hydrochloric acid is usually employed for dissolving out the mineral constituents of certain tissues, such as bone or teeth. As a rule, it is

better to use dilute acid (one of acid to three or four of water). A longer time must of course be allowed for the completion of the change than when the acid is concentrated.

Hydrochloric acid renders muscular tissue transparent. By a very weak solution of the acid in glycerine this object may be advantageously attained. Hydrochloric acid is also useful for softening and dissolving connective tissue, such as that between the gland follicles, muscular fibres, and nerve fibres. For these purposes a 1 per cent. solution is strong enough.

207. Chromic Acid.—Bichromate of Potash, &c.—Chromic acid is of great use to the observer for the purpose of hardening exceedingly soft and delicate tissues, and particularly nerve tissues of all animals. It was first employed by Hannover, in 1840. It has since been used by every microscopist, and is especially valuable for hardening sections of the brain, spinal cord, and ganglia. For hardening such tissues a solution containing from 5 to 1 per cent. is strong enough. Frey states that the best results are obtained by increasing the strength from time to time. Soft nerve organs, such as the retina, should be soaked in very weak solutions ($\frac{1}{2}$ per cent.) for some time, in order that they may be hardened very gradually. For the method of preparing the retina and the cochlea, see the second part of this work.

H. Müller suggested a fluid which contains bichromate of potash, and is very useful for some enquirers. Its composition is given on page 46.

Perchloride of iron, diluted with a large quantity of water, till of a very pale yellow colour, has been used by Führer and Billroth for hardening the spleen. Nitrate or silver has been used for staining the outer part of cells, as well as bioplasm (see page 69).

208. Effects of Alkalies.—The action of alkalies, even when cold in a very dilute state, is to soften, and at length to dissolve most animal textures. Cell-membranes, and many kinds of formed material, are quickly rendered extremely soft and transparent by alkalies, while the nucleus appears to be altered but slightly. Alkalies are also employed for dissolving certain crystalline substances which are occasionally found in animal tissues, such, for instance, as deposits of alkaline urates, which are not unfrequently met with in the form of considerable deposits in the tissues of gouty persons.

209. Potash and Soda.—The action of potash and soda upon animal structures is very similar. Both dissolve substances of an albuminous nature, but the effect of soda is more gradual, and it has been found that for most purposes in microscopical research, this reagent possesses advantages over potash.

The solution of potash is the ordinary *liquor potassæ* of the pharmacopœia, and the solution of soda is prepared in the same

manner. Both may be obtained perfectly pure of the operative chemists. These solutions may be diluted with from ten to twenty times their bulk of water. Potash and soda are employed where a tissue is to be rendered more transparent for the purpose of demonstrating the arrangement of the nerves or other anatomical elements not soluble in this reagent. The advantage of alkalies in studying the development of bone is referred to in page 48. See also plate V.

Potash soda, and, but to a less extent, ammonia dissolve the layer of epithelium covering mucous membranes, or render it perfectly transparent, so that the arrangement of the structures beneath can be easily demonstrated. In investigating the termination of the nerves and vessels in papillæ and other structures, they are very valuable.

For the purposes above mentioned, the alkalies should be diluted with water or glycerine. The changes are expedited by the application of a gentle heat—under 100°, or there will be danger of complete solution of all the tissues taking place. In the case of the structures, which are hard and dry, a higher temperature may be employed without danger. These may be warmed with the reagent in an ordinary test tube, as has been recommended by Kölliker.

Weak solution of ammonia, lime water, and baryta water have all been employed in the investigation of animal tissues. Lime water has been strongly recommended by Rollett in conducting investigations on the connective tissues, tendon, &c.

Carbonates of Potash and Soda.—Some animal textures become hardened by prolonged maceration in carbonate of potash, but this plan does not appear to be so generally useful as others previously indicated. Epidermic structures are not much altered by these salts. Gurlt recommends skin to be hardened in solution of carbonate of potash for the examination of the sweat ducts.

The introduction of different chemical solutions by injection, has been discussed in p. 95, and is the most efficient method of subjecting tissues to the action of reagents.

Of Obtaining Crystalline Substances from the Fluids and Textures of Animal Bodies.

Under this head it is proposed to give a sketch of a few of the simplest methods of obtaining certain crystalline bodies from animal solids and fluids. It is, however, inconsistent with the plan of this work, to attempt more than to allude to a few of the most important; and, for further information, the student is referred to the works enumerated in the note,* and particularly to the third volume of Dr. Miller's "Elements of Chemistry."

* "Lehmann's Physiological Chemistry," translated for the Cavendish Society; Gorup-Besanez, "Anleitung zur Zoochemischen Analyse;" Bowman's "Medical

210. Formation of Crystals in Animal Fluids.—Some crystalline bodies are deposited from their solution in animal fluids by simple evaporation; others, less soluble, may be deposited by allowing the fluid to stand still for a short time, when certain changes occur in some of its constituents, which lead to the precipitation of some bodies in a crystalline form, as, for instance, when uric acid or crystals of triple phosphate are precipitated from certain specimens of urine. In most cases, however, it is necessary to add some reagent before the crystals are thrown down, while not unfrequently a long and often complicated chemical analysis must be performed, if it is desired to isolate substances which were previously held in solution, and obtain them in a crystalline state. Even the addition of water will, in some cases, cause the most rapid crystallization, especially when the crystallizable material is dissolved in other media, contained in the interstices of tissue or is in a colloid state. Instead of water, it may be necessary to add alcohol, in which fluid the crystalline matter may be much less soluble.

Some crystalline bodies which are soluble at the temperature of the body, crystallize when the solutions containing them are cooled thirty or forty degrees. The effect of dilution upon retaining crystals in solution, need scarcely be alluded to.

Crystalline substances which are dissolved in animal fluids, may often be separated from them in a pure state by the addition of another fluid in which the crystals are not readily soluble, but which is readily miscible with the animal fluid. The fluid to be added should be introduced very gradually, in order that sufficient time may be allowed for the slow formation of the crystals, otherwise an amorphous precipitate may alone result. Some organic substances, soluble in alcohol, may be crystallized by the addition of ether to the alcoholic solution, while some are precipitated from their solution in water by the gradual addition of alcohol. Glycerine is a neutral substance which takes up water and thus assists to promote crystallization in many cases.

211. Influence of certain Constituents upon the Crystallization.—In many instances, it is exceedingly difficult to separate some crystalline bodies from other constituents with which they are retained in solution. Their solubility is perhaps increased, and their crystallization may be interfered with by the matters associated with them. The extractive matters of blood, urine, &c., exert this influence in a marked degree, and it is only of late years that several new bodies of definite chemical composition have been separated from extractives. Creatine and creatinine, leucine and tyrosine may be instanced amongst the number, for these were some time ago unknown and were included under the

Chemistry." Also the excellent "Lehrbuch der Zoochemie," by Heintz; the work of Hoppe-Seyler; Kühne, "Lehrbuch der Physiologischen Chemie;" and "Outlines of Physiological Chemistry," by Dr. C. H. Ralfe.

indefinite term "extractives." Certain colouring matters of definite composition have also been separated, and it is very probable that, as the methods of analysis at our disposal become improved, many new crystalline bodies will be discovered in the extractive matters. A very small quantity of extractive matter will entirely prevent the crystallization of urea, while the presence of chloride of sodium favours the separation of this material from solutions of organic matter by forming with it a compound which readily crystallizes in large octahedral crystals even in the presence of extractive matters.

The existence of carbonic acid in excess may cause carbonate of lime, triple phosphate, and other salts, to be held in solution. Excess of alkali will prevent the precipitation of uric acid, and excess of acid, that of phosphate of lime. Oily fats dissolve cholesterine, and serum possesses the power of retaining small quantities of both the latter substances in solution.

Hence, before the presence of many substances can be detected by microscopic examination, certain chemical operations are required in order to separate them from their solutions or combinations in the animal body, or for the removal of other substances which interfere with their crystallization.

212. Separation of Crystals from Animal Substances.—Not unfrequently, even after crystals have been obtained, if not very soon separated from the fluid in which they were formed, they again undergo solution or become decomposed. If the crystals are not very soluble, the supernatant fluid, or mother-liquor, may be poured off. The crystalline deposit is to be washed with ice-cold water, and subsequently dried on filtering paper or on a clean porous tile over sulphuric acid, without the application of heat. If the crystals will not bear the application of water, as much of the fluid as possible must be poured off, and the remainder absorbed with bibulous paper, or they may be placed upon a porous tile, and dried quickly over sulphuric acid *in vacuo*. In many instances we are enabled to wash the crystals with water, holding a little acid or alkali, or some alkaline salt, in solution, or with alcohol, chloroform, ether, or some other fluid in which we know them to be quite insoluble.

In some cases in which crystals insoluble in water are deposited in animal solids, they may be separated by agitation, when, being heavier than the water, they subside to the bottom, and the lighter animal matter may be removed by forceps, or if in a very minute state of division, poured off with the supernatant fluid. In other cases it may be separated by straining, the crystals being washed through the sieve which is usually made of fine muslin.

213. Examination of Crystals under the Microscope.—Some crystals which have been entirely separated from the fluid in which they

were originally deposited, may be examined in the dry way, in water, or other fluid in which they are known to be insoluble, or in Canada balsam; but, as a general rule, it is necessary to examine the crystals as they lie in some of the fluid in which they have been formed. When they have been obtained by allowing a concentrated solution to cool, some of the inspissated fluid must be removed with the crystals, placed upon a glass slide, or in a thin glass cell, covered with a piece of thin glass, and examined in the usual way—first using a low power (an inch), and afterwards a higher power (a quarter), because, although some of the crystals are of a large size, others amongst them, the form of which is very perfect, are often exceedingly minute. The crystals and mother-liquor should not be exposed to the air previous to examination, for in many instances water is absorbed, and partial solution takes place.

A drop of the solution should also be evaporated rapidly nearly to dryness, and allowed to crystallize upon the slide without being covered over, when the substance will often be found to assume a variety of beautiful forms, such as crosslets, dendritic expansions, &c., which vary according to the rapidity with which the evaporation has been conducted, and other circumstances, pl. XVII, figs. 1, 2.

214. Of obtaining Crystals for Examination.—In order to accustom himself to the necessary manipulation required in the process, the student may evaporate a solution of common salt upon a glass slide, and when it has become sufficiently concentrated, it may be covered with a small piece of thin glass, and allowed to cool. When cold it may be subjected to microscopical examination, and beautiful cubes of chloride of sodium will be observed, pl. XVII, figs. 1, 2. Crystals of several salts may be made in the same simple manner, and from an attentive examination of them much may be learnt. Solutions of phosphate of soda, phosphates of soda and ammonia, sulphates of potash and soda, muriate of ammonia, and a variety of other salts, may be readily made to yield microscopic crystals. Crystals which are precipitated by the addition of some reagent, such as nitrate of urea by nitric acid, must be examined in a little of the solution. The addition of water would, in many instances, destroy them immediately.

Different faces of the crystal, as it lies in the liquid, may be brought into view by slightly moving the thin glass cover with a fine-pointed instrument, such as a needle, while the preparation is in the field of the microscope. Octahedra of oxalate of lime found in urine may be turned over and over, while in the field of the microscope in this way. With a little practice, almost any crystals may in this manner be made to rotate in the mother liquor.

The influence of the crystals upon polarised light (H. to W. § 34), should be examined, and in cases in which the nature of the crystal has not been ascertained, its angles should be carefully measured, and

accurate drawings made. Its behaviour with chemical reagents is next to be ascertained, and the solubility of the crystals in water, alcohol, and other fluids should be noted. For these experiments different portions must be taken and separately tested in the manner referred to in §§ 200, 202.

215. Of Measuring the Angles of Crystals.—The goniometer is employed in the measurement of the angles of crystals. Although not much used in this country at present, it is important briefly to refer to its construction, as in some researches it is desirable that the angles of the crystals should be ascertained. Crystals which nearly resemble one another in their general form, and even in size, will be found to exhibit differences if their angles be carefully measured.

The simplest method of measuring the angles of microscopic crystals is that of Schmidt. The goniometer, pl. XXIV, fig. 4, consists of a positive eye-piece, which is so arranged as to be easily rotated within a large and accurately-graduated circle. Across the focus of the eye-piece a single cobweb is stretched; and to the upper part is attached a vernier. The crystals being placed in the field of the microscope, *and care being taken that they lie perfectly flat*, the vernier is brought to zero, and then the whole apparatus turned until the line becomes parallel with one face of the crystal; the framework bearing the cobweb with the vernier is now rotated until the cobweb becomes parallel with the next face of the crystal, and the number of degrees which it has traversed may then be accurately read off.

The cobweb goniometer just referred to will answer all the purposes for which such an instrument is required by the physiological or pathological observer, but for special crystallometrical investigations, a more elaborate apparatus becomes necessary. Dr. Leeson has applied the property of double refraction, possessed by Iceland spar, to the measurement of the angles of crystals under the microscope. A modification of his apparatus is described in "How to Work with the Microscope," (§ 68). See also a paper by Mr. Highley in the fourth volume of the Quarterly Journal of Microscopical Science, page 77.

216. Preservation of Crystals as Permanent Objects.—The preservation of the more soluble crystals is attended with the greatest difficulty, except when dried, in which state their microscopic characters are much altered. Crystals which very readily deliquesce on exposure to air, must be dried in vacuo, removed quickly to a cell, the cover of which must be firmly cemented down at once. Some crystals may, however, be dried and mounted in Canada balsam; others, such as oxalate of lime, cystine, triple phosphate, &c., can be well preserved in aqueous solutions, containing a little acid in the case of the two former substances, or an ammoniacal salt, in the latter instance, in which the crystals are known to be insoluble. Crystals which contain water of crystallization,

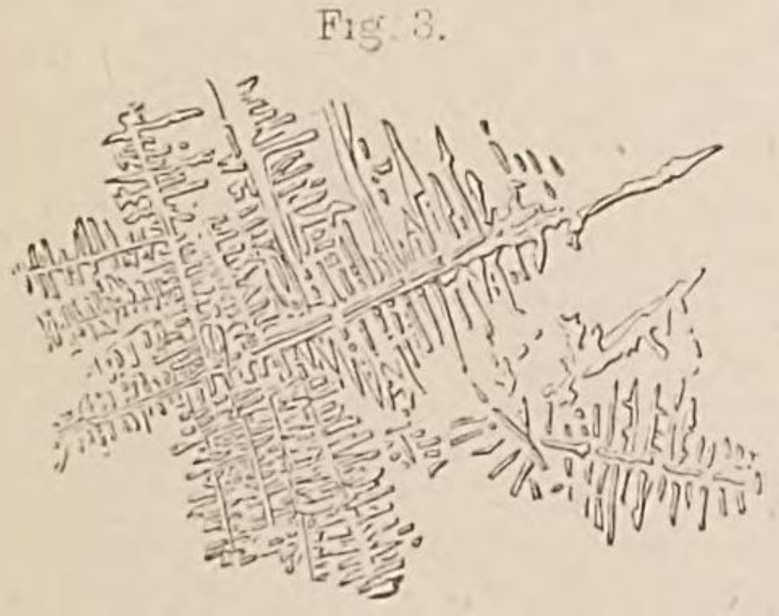
Fig. 1.

Chloride of sodium. × 215. p. 177.

Fig. 2.

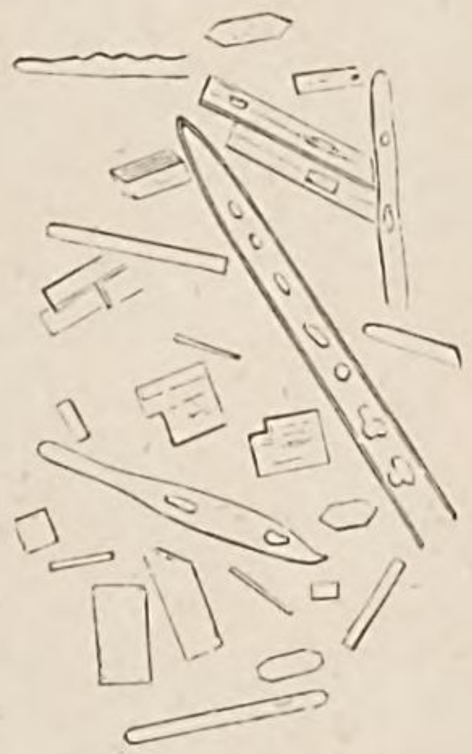
Chloride of sodium. × 215. p. 177.

Fig. 3.



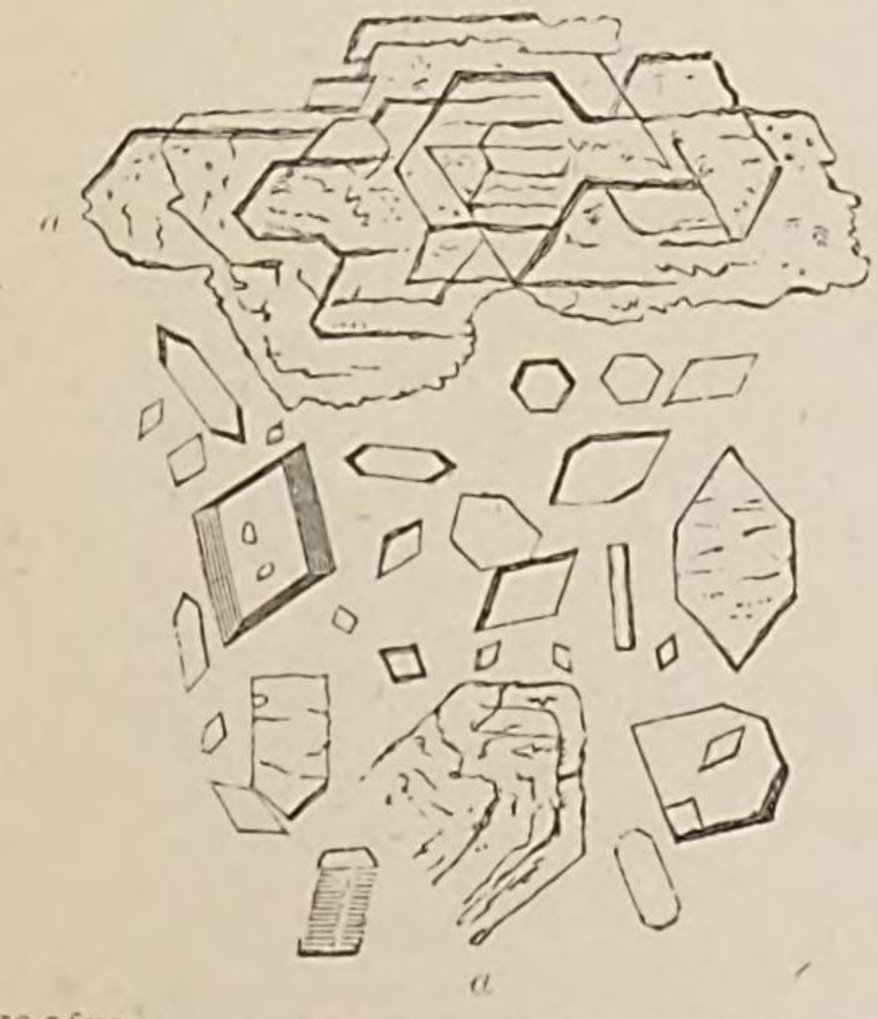
Chloride of ammonium. p. 177.

Fig. 4.



Urea. × 215. p. 179.

Fig. 5.



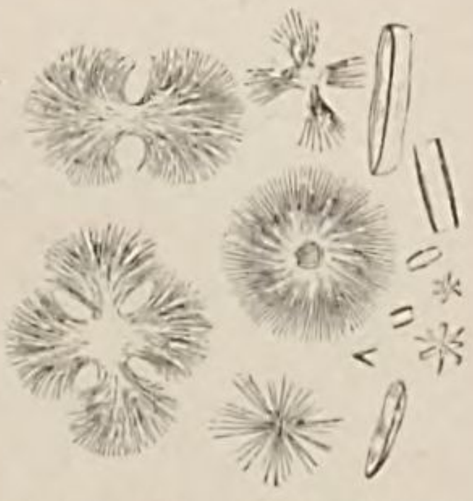
a

Nitrate of urea. *a*, crystals obtained from urine *b*, crystals of pure nitrate of urea × 215. p. 179.

Fig. 6.

Oxalate of urea obtained by adding oxalic acid to concentrated urine × 215. p. 179.

Fig. 7.



Wartz granules of the chloride of zinc, creatine, and creatinine, from urine. × 215. p. 181.

[To face page 178

may sometimes be preserved permanently in a drop of the mother-liquor; but, in many instances they alter much in form, and when we come to examine them, instead of finding a great number of small, well-formed crystals, as when the preparation was first put up, nothing remains but one or two large ill-shaped masses. The concentrated mother-liquor often acts upon the cement with which the glass cover is attached to the cell, or to the glass slide. After a time air enters, and the preparation is destroyed. Many crystals may be preserved in strong glycerine without much change taking place. I have some crystals of Guinea-pig's blood which have been preserved for several years in this medium, but some specimens mounted in the same way have quite failed.

A preparation of nitrate of urea in my possession has kept well for a considerable time in a very thin cell, containing only just sufficient of the mother-liquor to preserve the form of the crystals. The cell is made of Brunswick black. Crystals of chloride of sodium appear to keep pretty well in their mother-liquor, and the same will be found to be the case with a great number of substances. The more soluble crystals of an organic nature can seldom be preserved unless they are perfectly pure.

Of Crystals from Animal Fluids.

217. Urea.—Traces of urea in an animal fluid may always be detected by the crystalline characters of the nitrate of urea. Upon adding a drop of nitric acid to a drop of cold concentrated urine, or other solution containing urea, placed upon a glass slide, a crystalline precipitate of nitrate of urea will immediately take place. Upon covering this with a piece of thin glass, and subjecting it to microscopical examination, the characteristic rhomboidal plates will be observed. Fig. 5, pl. XVII, represents the appearance of nitrate of urea examined with a quarter of an inch object-glass, at *a* are shown some crystals of the impure nitrate, as obtained from urine; the other crystals in the figure were formed by adding some nitric acid to a solution of pure urea.

Another drop of the concentrated urine may be treated with a strong solution of oxalic acid, when we shall obtain crystals of oxalate of urea, the form of which is represented in fig. 6, under a quarter of an inch object-glass. When mere traces are suspected to exist in animal fluids or solids, we must proceed to separate the urea from albuminous or other substances, before the addition of the nitric acid. If the urea exist in an albuminous solution (serum of blood, or in a dropsical fluid), we must remove the albumen by boiling with a few drops of acetic acid, and subsequent filtration. The filtered solution is to be evaporated to dryness over a water-bath, and the dry residue treated with cold acohol. As a general rule, however, I think it preferable to evaporate the solution supposed to contain urea, at the temperature of 100°, or in vacuo and treat the dry residue with alcohol, which dissolves the urea. Much

chloride of sodium separates from the alcoholic solution as it is evaporated. If to a little of the cold mother-liquor a drop of nitric acid be added, as above described, crystals of nitrate of urea will be formed, if urea was present in the original solution. In all cases, the fluid suspected to contain urea must be operated upon when quite fresh, as this substance readily becomes decomposed into carbonate of ammonia. Numerous well-formed crystals of urea, oxalate of urea, and nitrate of urea, are figured in the plates of my work on "Kidney Diseases, Urinary Deposits, and Calculous Disorders."

In examining solid organs for urea, the fresh tissue may be broken up, and treated with several portions of hot alcohol, the solution filtered from coagulated matters, and evaporated. The residue may again be extracted with alcohol, and the pure crystals may be obtained by solution in water and subsequent evaporation.

Oxalate of Urea is easily prepared by adding crystals of oxalic acid to a concentrated solution of urea, or to urine evaporated to the consistence of syrup. As the mixture becomes cold, numerous crystals of oxalate of urea form, pl. XVII, fig. 6.

Crystals of pure urea, obtained by decomposing a solution of the oxalate of urea with chalk, and carefully evaporating the filtered liquid, are shown in fig. 4. The cavities represented in many of the crystals contain fluid. Urea may also be obtained in a nearly pure form by adding ether, in which it is only slightly soluble, to the fluid which contains it.* Urea may be determined, quantitatively, by weighing the nitrate and calculating the proportion of urea it contains, by decomposing it with solution of chlorinated soda and estimating the volume of nitrogen according to the method of Dr. Davy.† or by Liebig's process.‡

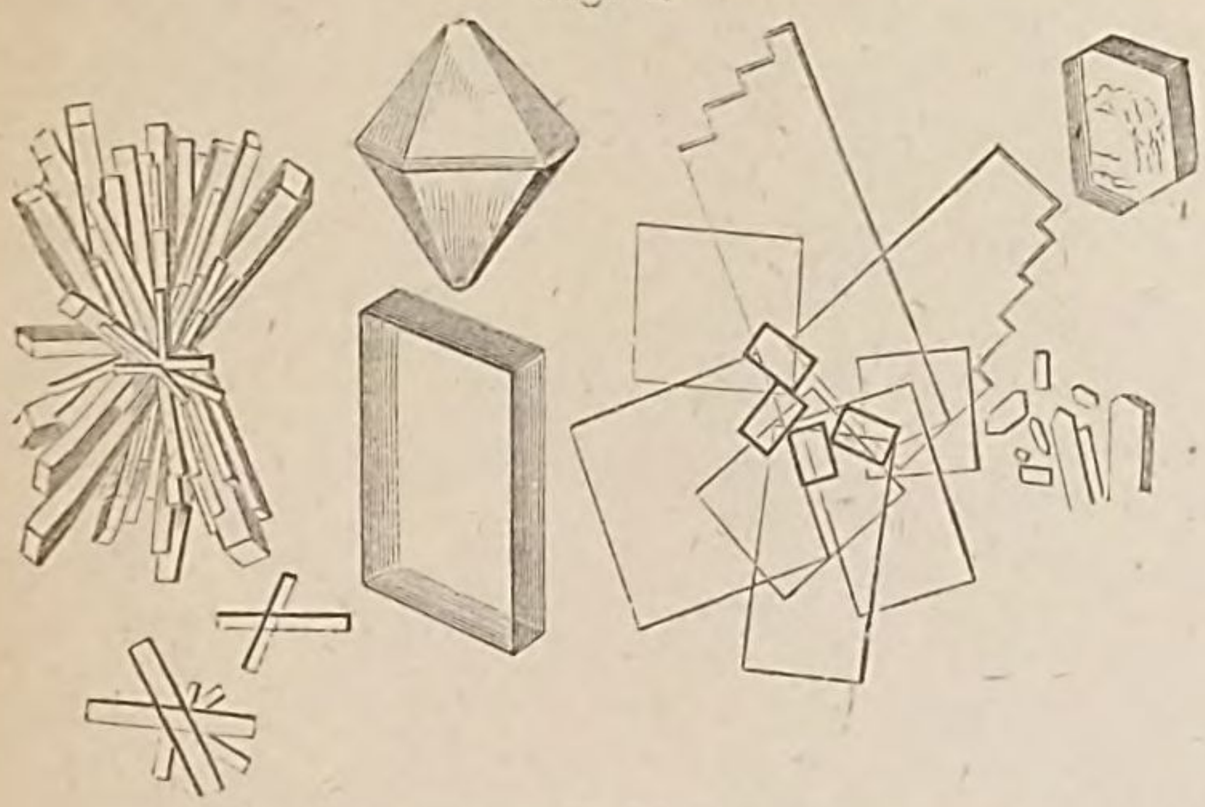
218. Creatine—Creatinine.—Creatine exists in very small quantity in muscular fibre. Traces of it are also present in urine, in which fluid it was discovered by Liebig. According to Dr. Gregory, it is most readily prepared from the flesh of the cod fish; from twenty-five pounds of which, in one experiment, he obtained 164 grains of creatine. From crocodile's fresh I obtained it very readily; two pounds yielded more than seventeen grains of pure creatine. The flesh is to be chopped in small pieces, and well kneaded with water. After all the fluid has been expressed by powerful pressure, it is very carefully raised to the boiling-point, and the coagulated matter removed by filtration. The phosphatic salts are precipitated by caustic baryta. The solution must be again filtered, and evaporated at a gentle heat (130°–140°) to about one-twentieth of its volume, or to the consistence of syrup; any scum which

* A plan recommended by Dr. Marcet.

† "Dublin Hospital Gazette," June 1st, 1855; "Archives of Medicine," vol. i, page 144; "Kidney Diseases, Urinary Deposits, and Calculous Disorders." 3rd edition.

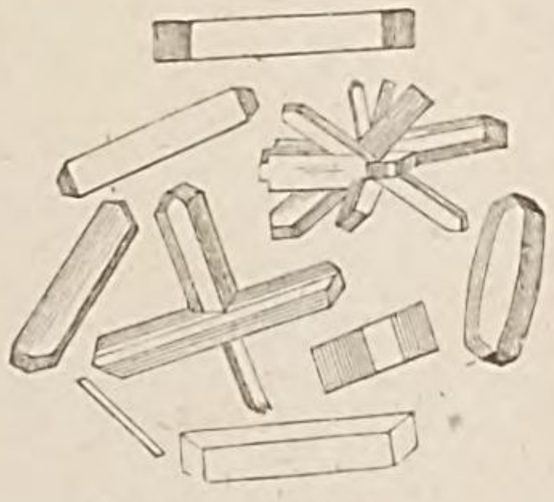
‡ Vide a paper by Dr. von Bose, "Archives of Medicine," vol. i, page 34.

Fig. 1.



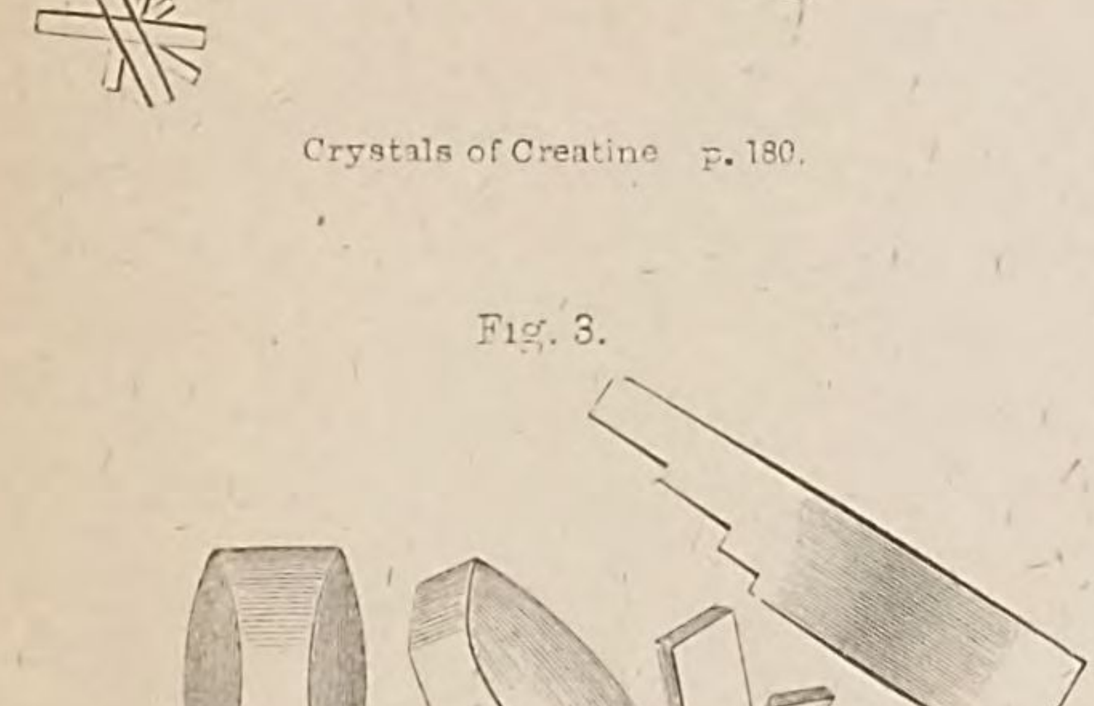
Crystals of Creatine p. 180.

Fig. 2.



Creatine. p. 181.

Fig. 3.

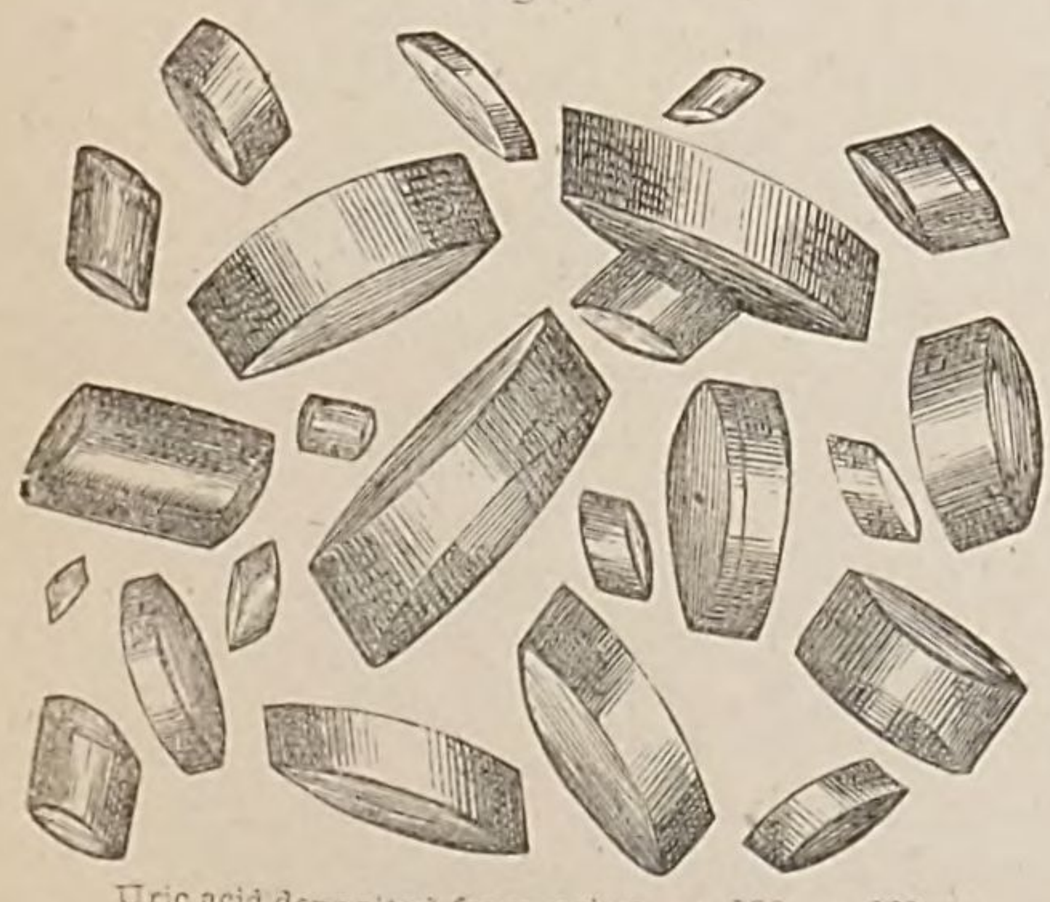


Crystals of Creatinine. p. 181.

Fig. 4.

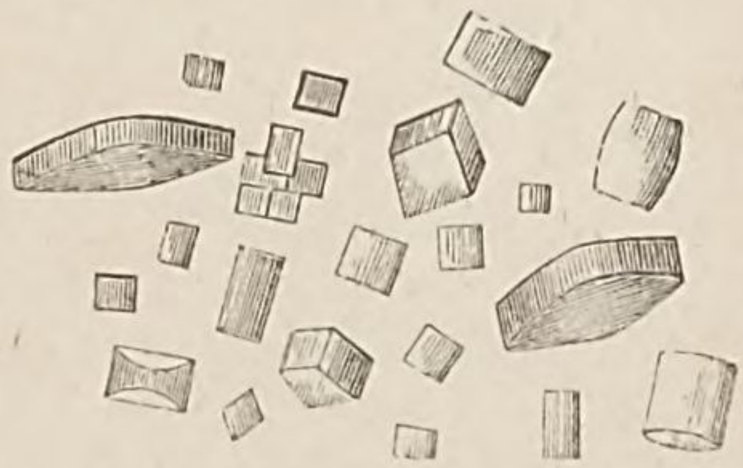
Uric acid, deposited from alkaline urates by the addition of excess of acid × 215 p. 181.

Fig. 5.



Uric acid deposited from urine. × 180. p. 181

Fig. 6.



Uric acid from urine. × 215. p. 181.

Fig. 7.

Rare form of uric acid crystals deposited from urine. × 215. p 181.

[To face page 180.

forms from time to time is to be removed from the surface. This concentrated solution may then be set aside. On cooling, it forms a thin jelly, and, after standing for some days, crystals of creatine are deposited.

Crystals of creatine are represented in pl. XVIII, figs. 1 and 2, and those of creatinine in fig. 3. The first and last have been copied from the Atlas of Robin and Verdeil.

I have obtained crystals of creatine and creatinine from urine, according to Liebig's process, as follows:—A quantity of urine was neutralized by lime water and precipitated by chloride of calcium. The filtered solution, after being evaporated to a small bulk, was again filtered from the saline residue which crystallized out, and mixed with about one-fourth of its weight of a solution of chloride of zinc, previously concentrated to a syrupy consistence. After some days had passed, numerous warty masses of a compound of chloride of zinc and creatinine, with which the creatine was mixed, separated, pl. XVII, fig. 7. These were re-dissolved in water and crystallized. The pure crystals were boiled in water with hydrated oxide of lead, and the chloride of lead and oxide of zinc separated by filtration. The solution containing the creatine and creatinine was concentrated. The crystals thus obtained were purified by re-crystallization, and treated with boiling alcohol, which dissolved the creatinine, leaving the creatine behind. By purification with animal charcoal and re-crystallization, excellent crystals were obtained. Dr. Von Bose, obtained a considerable quantity of these crystals from urine in my laboratory (1856).

219. Uric or Lithic Acid.—The presence of uric acid in a crystalline form, can be readily detected in animal fluids and solids, by microscopical examination, if it occur in a crystallized state.

In order to ascertain if an amorphous or other deposit contain uric acid, or a urate, we must treat it with a few drops of potash, which will dissolve any of the acid that may be present. This alkaline solution is to be decomposed with excess of acetic acid, and, after the mixture has been allowed to stand for twenty-four hours or longer, any deposit that may have formed, is to be subjected to microscopical examination. The microscopic crystals of uric acid, obtained in this manner, are usually in the form of rhombic tablets, pl. XVIII, figs. 5, 6, and 7, but sometimes they assume the form of six-sided plates. Uric acid not unfrequently crystallizes from urine, without the addition of any free acid. The crystals vary extremely in shape. Common and rare forms are represented in the figures. See also "Kidney Diseases, Urinary Deposits, and Calculous Disorders," in which work more than twenty different crystalline forms are given.

Uric acid is soluble in alkaline fluids, and when present in serum, exists in combination with an alkali. If uric acid or a urate existed in a fluid, we should be able to detect it in the aqueous extract without

difficulty. It is only necessary to concentrate the solution, and then add excess of acetic acid.

Dr. Garrod (Medico-Chirurgical Transactions, vol. xxxi), has proposed an excellent plan for detecting the presence of uric acid in the blood of gouty patients, which is very simple and easy of execution. A little of the serum is poured into a watch-glass, and a few drops of acetic acid, which does not coagulate albumen, added to it. Two or three very fine filaments of silk, or tow, are then placed in the mixture, and the whole allowed to stand in a still place, under a glass shade, for twenty-four hours or longer. Upon submitting the filaments of tow to microscopical examination, they will often be found studded with minute crystals of uric acid, frequently exhibiting some of the forms shown in the above figures.

The student will gain much practical information as to the characters and various forms which this substance assumes, by dissolving some of the crystals obtained from urine in alkaline solutions (potash, soda, alkaline carbonates, phosphates, &c.), and then causing the crystals of uric acid to be precipitated by the addition of excess of acid. To some specimens he may add hydrochloric, to others, acetic or nitric acids, &c. Upon examining in the microscope the crystals obtained by these different means, he will notice a great variety of forms, but, upon careful examination, it will be found that most of them are mere modifications of the same form, and that a gradation between some of them may be traced in certain instances. See the chapter on Urinary Deposits, and my work on "Kidney Diseases, Urinary Deposits, and Calculous Disorders."

220. Hippuric Acid is separated from its salts by the addition of a stronger acid (as hydrochloric acid) and the crystals which are deposited may be subjected to microscopical examination in the usual manner. Hippuric acid should always be sought for in animal fluids which are quite fresh, as it undergoes decomposition very rapidly, and becomes entirely converted into benzoic acid. The microscopical characters of these two acids are, however, very distinct. Benzoic acid crystallizes in scales, while the crystals of hippuric acid occur in the form of beautiful prisms, pl. XIX, fig. 1, not unlike those of the ammoniaco-magnesian phosphate. Hippuric acid is very soluble in hot water, and also in alcohol. Solutions of hippuric acid redden litmus paper strongly.

In order to detect small quantities of hippuric acid, the animal fluid supposed to contain it must be perfectly fresh. It is to be evaporated nearly to dryness, and then treated with alcohol, sp. gr. ·830. After the addition of a crystal of oxalic acid, the spirituous solution is evaporated to the consistence of syrup. The residue is next to be extracted with ether, which contains about one-sixth of its volume of alcohol. The solution is again evaporated, and the remaining extract treated with

water, which dissolves the hippuric acid, while any fatty matter which is present is left behind in an insoluble state. The solution may be filtered into a watch-glass, and allowed to evaporate slowly that crystals may form.

Hippuric acid may always be obtained from the fresh urine of horses or oxen. After the administration of benzoic acid, it may be found in human urine, as was demonstrated many years ago by the late Mr. Ure; and Lehmann has remarked the presence of hippuric acid in diabetic urine, in every instance in which he has sought for it. Lehmann states, that in diabetic urine, hippuric acid takes the place of the uric, which, according to him, is absent in this condition. Some exception to this statement must, however, be taken, for I have seen many cases of confirmed diabetes in which the urine not only contained uric acid, but in which this substance was present in very large quantity and formed an abundant deposit. The fact has been observed by other practitioners in this country, and probably years before Lehmann called attention to the matter. Indeed, in this country at least, I am quite certain that uric acid is often found in diabetic urine. Hippuric acid has been found in the blood of oxen by Verdeil and Dolfuss.*

221. Lactic Acid.—Lactates.—The presence of this acid is often detected with difficulty in animal substances, in consequence of its characteristic reactions being interfered with by the presence of many organic bodies. Its separation from other substances is attended with much trouble, especially when it is present only in very minute proportions.

It has long been supposed that rheumatism is due to an excessive production of lactic acid, or to its undue accumulation in the blood. Dr. Richardson states that he introduced a rheumatic condition in dogs, accompanied by affection of the joints and heart, by injecting a dilute solution of lactic acid into the peritoneal cavity. The presence of lactic acid is most readily determined by the microscopical characters of certain of its crystalline salts. Of these the *lactates of zinc*, *copper*, *and lime* are the most characteristic.

In order to detect the presence of lactic acid in animal fluids, Lehmann proceeds as follows: the fluid is evaporated carefully over a water-bath, and the residue extracted with alcohol. After the separation of some of the salts by evaporating this alcoholic solution, and allowing them to crystallize out, the remaining mother-liquor is treated with sulphuric or oxalic acid. The sulphate or oxalate of potash is precipitated by means of alcohol, and the impure lactic acid remains in solution. To this solution baryta water is next added. The excess of baryta is removed by carbonic acid. The solution filtered from the

* Lehmann's "Physiological Chemistry," Cavendish Society, vol. ii, page 212.

precipitate is evaporated to a syrupy consistence, treated with alcohol, filtered, again evaporated, and then allowed to stand for some time, in order that any baryta salts may crystallize out. The syrup is next removed, and decomposed with sulphate of lime. The solution filtered from the sulphate of baryta is evaporated to a small bulk, when crystals of lactate of lime, in the form of double brushes, pl. XIX, fig. 2, with crystals of sulphate of lime, may be observed upon microscopical examination. The crystals of lactate of lime may be dissolved in alcohol and sulphate of copper added. After the removal of the excess of sulphates of lime and copper by evaporation and crystallization, the remaining solution is to be concentrated, and the crystals of lactate of copper examined in the microscope, pl. XIX, fig. 3. If distinct and measurable crystals are not obtained in this manner, Lehmann dissolves the residue in a little water to separate any butyric acid that may be present, and after being strongly boiled, the solution is filtered, and a zinc bar placed in it, which in the course of a short time, becomes covered with crystals of lactate of zinc, the angles of which may be measured with the goniometer, pl. XIX, fig. 4.

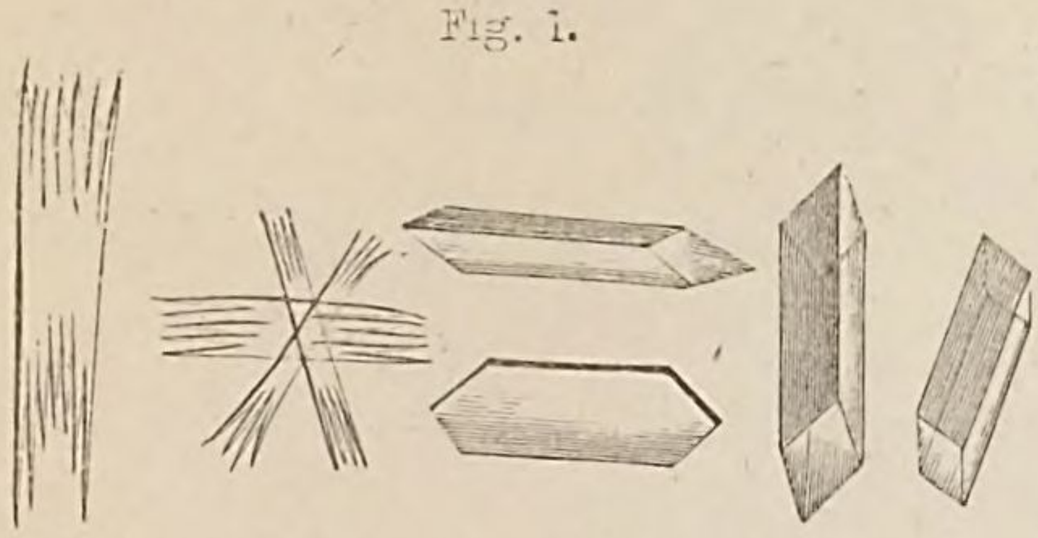
222. Taurine.—Pulmonic, or Pneumic Acid.—*Taurine* is obtained in the form of beautiful six-sided prismatic crystals, by decomposing bile with hydrocoloric acid. The bile is to be boiled with the acid for several hours. After filtration and evaporation taurine crystallizes out. This substance is found in the contents of the intestine, and commonly in the fæces. It has been detected in the urine in jaundice. Taurine is represented in pl. XIX, fig. 5.

The so-called pulmonic acid was discovered in the lung tissue, by Verdeil. It is prepared as follows: perfectly fresh calves' lung is cut into small pieces, and extracted with tepid water. It is well pressed, in order to remove all the liquid. The fluid is treated with sulphate of copper to precipitate the albumen. The excess of sulphate of copper is removed by the addition of sulphuret of barium, or by adding baryta water and passing sulphuretted hydrogen through the liquid. The filtered solution is evaporated to the consistence of syrup, and time allowed for the formation of crystals. These may be re-crystallized from spirit, to which a few drops of sulphuric acid have been added. In this manner the crystals represented in pl. XIX, fig. 7, were obtained. Some crystals obtained by myself according to these directions are represented in fig. 6. It is probable, however, that these do not consist of one simple substance. The so-called pulmonic acid of Verdeil has been considered by Cloëtta and others to be composed of taurine. Consult Scherer's memoir on "Hypoxanthine, Ann. der Chemie und Pharmacie," cxii, p. 257, and Stricker's paper on Sarcine, "Quar. Jour. Chem. Soc.," vol. x, p. 121, July 1857.

223. Leucine has of late been found in many of the solids and fluids

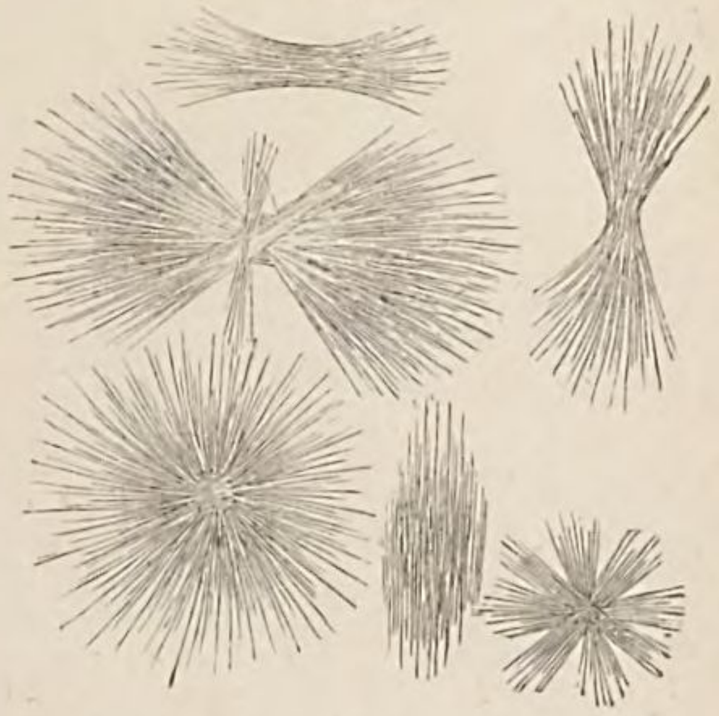
PLATE XIX.

Fig. 1.



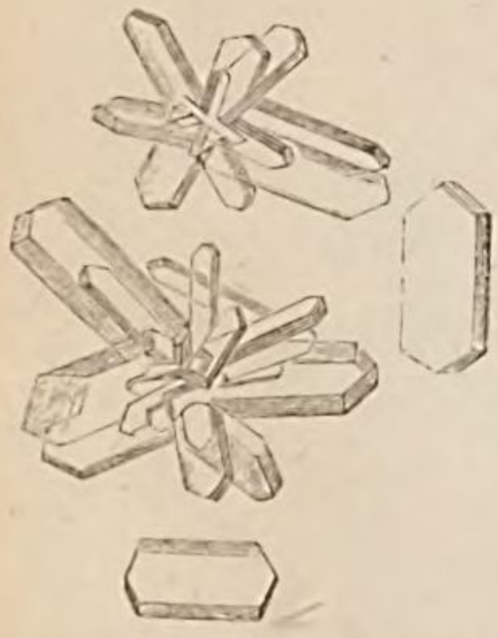
Hippuric acid. Robin and Verdeil. p. 182.

Fig. 2.



Crystals of lactate of lime. p. 183.

Fig. 3.



Lactate of copper p. 181.

Fig. 5.

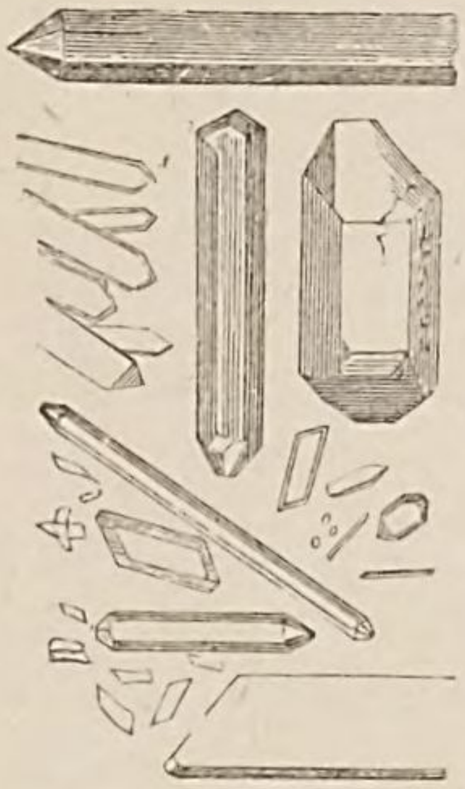
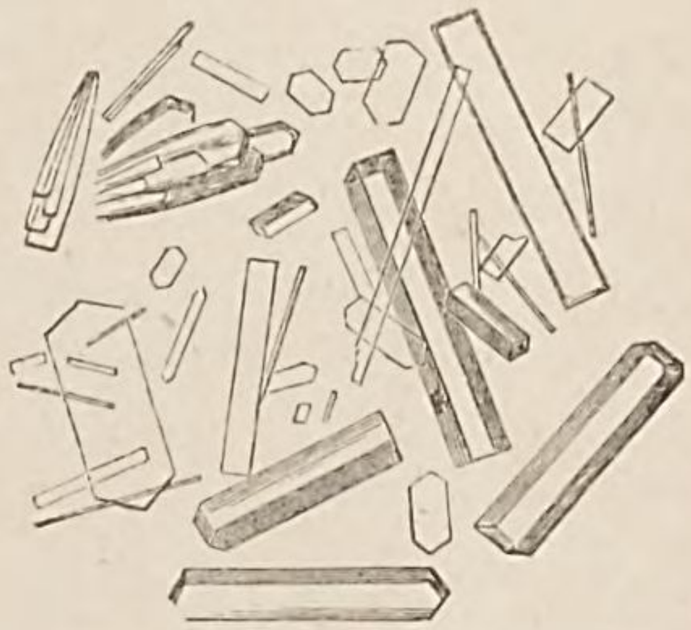


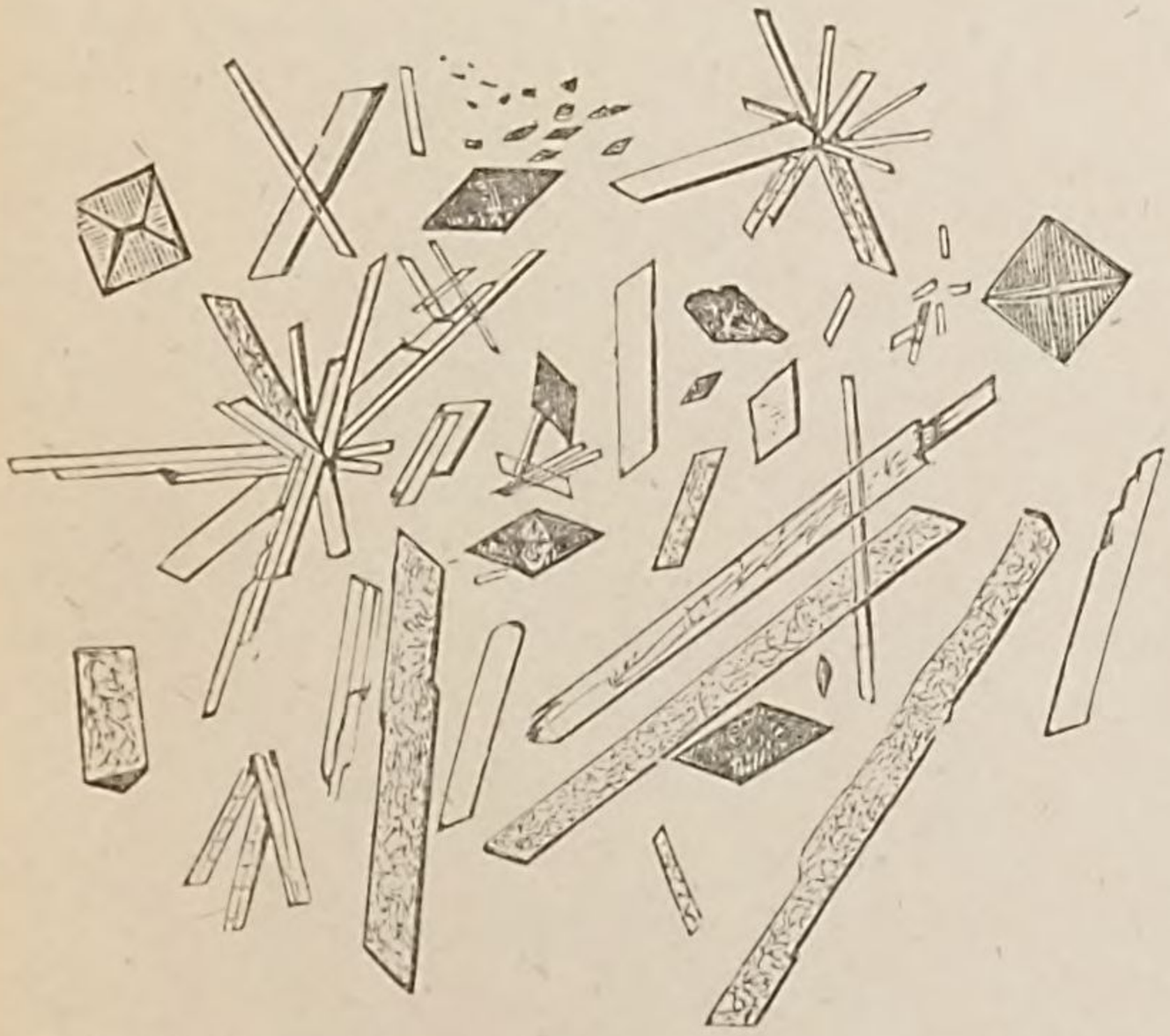
Fig. 4.



Lactate of zinc. p. 184.

Crystals of taurine. p. 181

Fig. 6.



Crystals from Cat's lung, the so-called pulmonic acid, probably taurine. × 215. p. 181.

Fig. 7.

The so-called pulmonic acid, after Verdeil. Probably taurine. p. 184.

[To face page 181.

of the animal body. It is not very soluble in water (one part in twenty-seven), but more so in alcohol. It crystallizes from aqueous solutions, for the most part in spherical masses, which exhibit a radiated arrangement, pl. XX, figs. 1 and 2. From alcohol leucine is deposited in the form of pearly scales, somewhat resembling cholesterine. Dry leucine can be sublimed without change. Leucine has been found in the saliva, pancreatic juice, and in the pulmonary tissue of the ox (Cloëtta*). Frerichs and Städeler have detected leucine in the blood, urine, and bile of patients suffering from typhus, small-pox, and other exanthemata. Scherer obtained six ounces of pure leucine from twenty pounds of ox pancreas, as well as fifteen grains of guanin and thirty of xanthin. Dr. Thudichum found leucine in the urine of a man, whose liver yielded a large quantity of it.† It was obtained by concentrating the urine. This substance is probably formed in the liver, and in health rapidly converted into other compounds. In certain diseases it is to be detected in very considerable quantity. Crystals of leucine may often be seen in sections of livers of persons who have died of jaundice. Frerichs has given several figures of leucine crystals in the liver and also in the urine. It occurs especially in the urine of patients suffering from acute yellow atrophy of the liver.‡

No satisfactory tests for leucine are yet known. If it can be obtained pretty pure by repeated recrystallization, the dried leucine may be sublimed. The sublimate of aggregations of rhombic plates could hardly be mistaken for anything else. Urate of soda and many other substances crystallize in spherical globes like leucine. Crystals of this form, however, which are soluble in alcohol and again crystallize in spherules, from an aqueous solution, can hardly be anything but leucine. This substance cannot, therefore, be recognized by the form of the crystals alone.

Leucine may be obtained in quantity by allowing cheese, albumen, or flesh to decompose with about fifty parts of water for six weeks. Decomposed liver yields a large quantity. The fluid is to be boiled with milk of lime. After precipitation of the lime by the cautious addition of sulphuric acid, the filtered solution is treated with acetate of lead. After filtration, the solution is evaporated to the consistence of syrup, and the leucine crystallizes out. The addition of alcohol favours the separation of the leucine. Lastly; it is dissolved in water, treated with sulphuretted hydrogen, and the leucine obtained pure by recrystallization. The best plan for obtaining leucine is to fuse yellow elastic

* "Chemical Gazette," 1856, page 61.

† "A Treatise on the Pathology of the Urine," 1858.

‡ "Pathologisch Anatomischer Atlas zur Leberkrankheiten," von Dr. Fried. Theo. Frerichs, Braunschweig, 1858. See also Dr. G. Harley's work "On Jaundice."

tissue, horn, wool, epithelium, or white of egg, with an equal weight of hydrate of potash. As soon as hydrogen begins to be evolved and the dark brown mass changes to a yellow colour, it is removed from the fire. The mass is treated with hot water and the highly alkaline solution is to be slightly supersaturated with acetic acid. Tyrosine first crystallizes by evaporation, but the leucine may be obtained by concentrating the mother-liquor. It may be purified as before mentioned.

In order to obtain leucine from urine, Frerichs recommends that, after concentration, the fluid should be digested with cold absolute alcohol. The extractives are gradually dissolved out. Upon treating the residue with boiling spirit of wine, the leucine is dissolved out, and crystallizes as the solution cools. It may be purified by recrystallization. If leucine is present in considerable quantity, it readily crystallizes out if the urine be concentrated to the consistence of syrup, and allowed to stand for a week or ten days. In this simple way I have often obtained crystals of leucine.

224. Tyrosine crystallizes in long white needles, which are often aggregated to form brush-like masses, pl. XX, fig. 3. It is hardly soluble in cold water, but it is readily dissolved by alcohol, ether, boiling water, the mineral acids, and alkalies. Tyrosine is probably formed in the liver with leucine. It has been detected in the urine of persons suffering from typhus fever, by Frerichs and Städeler. It has been detected in many of the animal fluids. Tyrosine may be obtained from pancreas and from albuminous matters by boiling with weak sulphuric acid or by boiling horn, feathers, or hair, with sulphuric acid and water for forty hours. The dark brown liquid is to be made alkaline with milk of lime, warmed, and then filtered. Sulphuric acid is added to neutralization, and crystals of tyrosine are deposited upon evaporating the liquid.

A very delicate test for this substance has been proposed by Hoffman. A solution of nitrate of protoxide of mercury, nearly neutral, is to be added to the solution suspected to contain tyrosine. If this body be present, a reddish precipitate is produced, and the supernatant fluid is of a very dark rose colour. Frerichs' test for tyrosine is as follows:— The matter supposed to be tyrosine is mixed with sulphuric acid in a small capsule. After the lapse of half an hour water is added. The solution is then boiled, and excess of carbonate of lime added. To the filtered solution a few drops of a solution of perchloride of iron which is free from acid is added. A dark purple colour is produced if tyrosine is present.

In order to obtain tyrosine from urine it is necessary to add a solution of acetate of lead until a precipitate is no longer produced. Sulphuretted hydrogen is passed through the filtered liquid. The sulphuret of lead being separated by filtration, the clear solution may be

concentrated by evaporation, when tyrosine, if present, will crystallize out.

De la Rue found tyrosine in the cochineal insect. This is doubtless one of the substances resulting from the disintegration of albuminous substances. I have found it in considerable quantity in urine which contained much uric acid, and had been left to stand in a warm place for many weeks. Leucine and tyrosine were detected by Dr. G. Harley, in the urine of a dog four days after dog's bile had been injected under the skin.—(*On Jaundice*, p. 96.)

225. Fatty Matters.—Some fats crystallize in characteristic forms, from their ethereal or alcoholic solutions.

Margarine may be readily obtained from human fat; it is deposited from its alcoholic solution in round spherical masses, which being composed of dense aggregations of minute crystals appear almost black by transmitted light, pl. XX, fig. 4. Almost the whole of the oily fat remains in solution in the alcohol. Heintz has proved that margarine really consists of a mixture of palmitine with stearine.

Minute stellæ of this substance may be obtained from a concentrated alcoholic solution of human fat. Not unfrequently crystals separate spontaneously from the oily fat in which they have been previously dissolved. This crystallization may sometimes be seen in the contents of the fat vesicle of specimens of adipose tissue, particularly if putrefaction has commenced, and also in many mixed fatty matters which have been extracted from animal substances. In certain cases dark colouring matters are taken up by the crystalline portion of the fat only. For example, in cases of cholera I found that the adipose tissue in the submucous areolar tissue of the small intestine exhibited dark reddish-brown stellæ upon the surface of the oily fat, producing a very beautiful appearance. The colour was derived from the altered and dissolved blood colouring matter, which had permeated the vessels and had been taken up in large proportion by the crystallizable fat of each vesicle. I have seen the same appearances in cattle plague and in cases of typhoid fever in the human subject.

The so-called margarine crystallizes from its solutions in tufts composed of somewhat wavy, minute, acicular crystals, or in separate, free, short crystals, which are usually somewhat curved. The so-called margaric acid also crystallizes in minute tufts composed of very small and much-curved crystals.

Stearine may be obtained in large quantity from mutton fat; it is only slightly soluble in hot alcohol, from which solution it readily crystallizes in a form much resembling that of margarine, but the needle-like crystals are for the most part thinner, and their direction is straight. Stearine also very commonly crystallizes in quadrangular tablets.

The characters of stearic acid under the microscope are shown in pl. XX, fig. 6, and those of the so-called margaric acid in fig. 7. These figures were taken from the excellent atlas of plates by Robin and Verdeil, "Traité de Chimie Anatomique et Physiologique," a work that may be consulted with great advantage by all interested in the microscopical characters of the various crystalline substances met with in, or obtained from, the animal body. Crystalline fatty matters are not unfrequently found in morbid growths, and very commonly in various fluids and solids of the body. In vomited matters, masses of crystalline fat are very often observed, and in vomit containing sarcinæ, stellar crytalline fatty matters are almost invariably present.

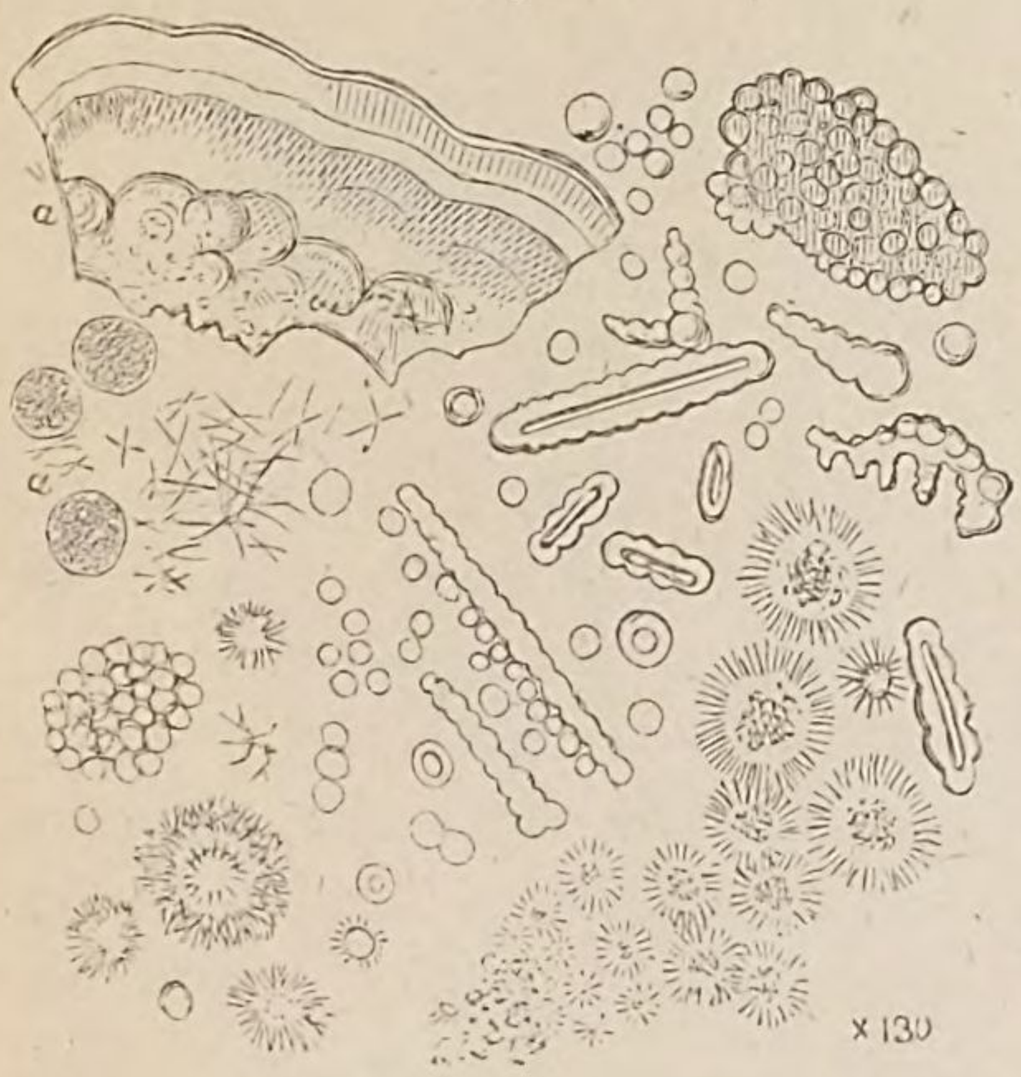
Oil globules are always present with the crystals of fats and fatty acids, and among the crystalline fatty matters, deposited from ethereal or alcoholic solutions, obtained by digesting various dried animal substances in alcohol or ether, a large number of oily fat globules will also be observed.

226. Cholesterine is a non-saponifiable fat, and occurs in many situations in the human body. In the tissues of the aged it is often present in considerable quantity. It is hardly to be detected in young textures but the amount increases as age advances, and this fact is particularly noticed in the case of nerve tissues. A small quantity of cholesterine is always present in bile, and the colourless gall-stones consist almost entirely of this substance. It may be extracted from many of the tissues in a state of health; I have even obtained it from the healthy crystalline lens of the eye.

In disease, cholesterine often occurs in serous fluids, especially in the serum of ovarian and other serous cysts, and occasionally in the fluid of hydrocele. It is present in many tissues in a state of fatty degeneration, and may be even extracted from the epithelial cells of the air passages in bronchitis. I have obtained it also from the epithelium and oil globules in the casts of the uriniferous tubes in fatty degeneration of the kidney. It has been often stated that cholesterine is never present in the urine, but I believe it is to be invariably detected in cases where oil casts exist in sufficient quantity for analysis.

Cholesterine may always be recognized by its crystalline form, but the angles of the crystals vary considerably in different cases, pl. XXI, fig. 1, and may usually be obtained by the slow evaporation of alcoholic solutions; but where only mere traces of this substance are present, it is necessary to remove the other fatty matters before the cholesterine can be obtained in the crystalline state. By boiling with water and oxide of lead, the saponifiable fats form a plaster. Cholesterine is dissolved by treating matter supposed to contain it with dilute alcohol, from which solution it may be obtained in a crystalline form by subsequent evaporation.

Fig. 1.



Crystals of Leucine. *a* crystallised from water, the remainder from an alcoholic solution. p. 185.

Fig. 2.

Crystals of Leucine. p. 185.

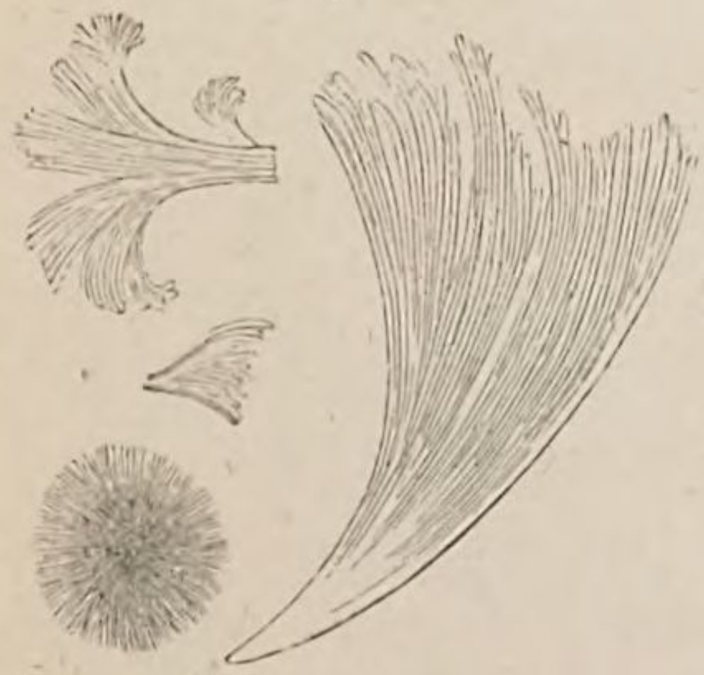
Fig. 3.

Tyrosine, crystallised from water. p. 186.

Fig. 4.

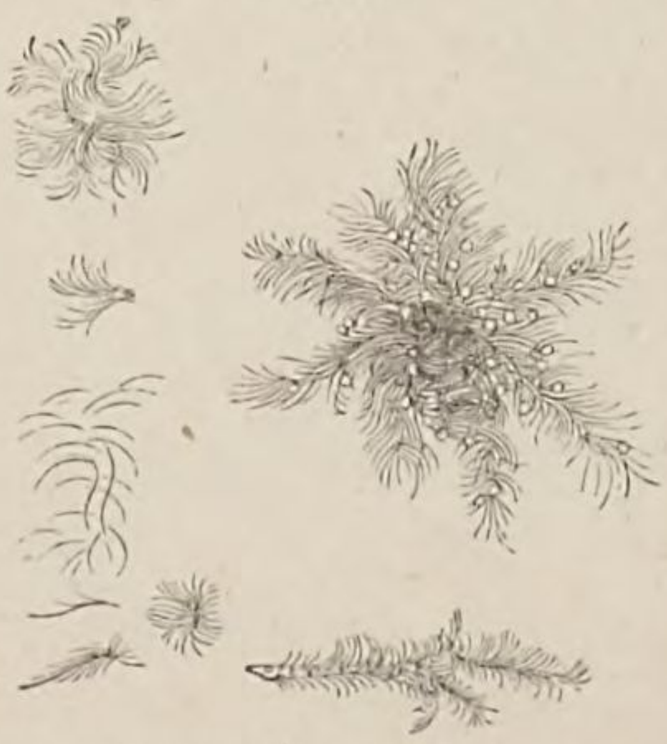
Crystals of Margarine, after Robin and Verdeil. p. 187.

Fig. 6.



Stearic acid. Robin and Verdeil. p. 188.

Fig. 7.



Margaric acid. p. 188.

Cholesterine is coloured of a dark red colour by the action of sulphuric acid.

Seroline.—This is another non-saponifiable fat discovered by Boudet in serum, but differs from cholesterine in not forming distinct and well-defined crystals; it separates in large transparent flakes from alcoholic solutions, fig. 2.

227. Myelin though not crystalline may be conveniently referred to this place. It is a colourless glistening semi-fluid substance prone to form drops, and capable of being drawn out into long threads which curve and twist into the most peculiar forms. If the observer examines a portion of the white matter of the brain or spinal cord in water, he will recognise this substance without difficulty. The masses often exhibit double contours, and not unfrequently many lines may be discerned equidistant from one another, but varying much in thickness and intensity, pl. XXI, fig. 6, *a*. It is present in the liver, and may be detected in almost all the tissues. In many tissues of the adult myelin exists in considerable quantity in the outer part of cells, fig. 4, and in old age a still larger proportion is present.

This substance was first described by Virchow, but Beneke has shown that it may be obtained from all the tissues of the body, and that it exists even in plants. It is soluble in hot alcohol, ether, and turpentine. Cholesterine is a necessary constituent of myelin, and can always be obtained from it.

Iodine tinges myelin of a reddish-brown colour. If sulphuric acid be added, a blue or violet colour is induced. This reaction probably depends upon the presence of cholesterine. Beneke showed that myelin gave the reaction characteristic of the biliary acids, upon the application of Pettenkofer's test.

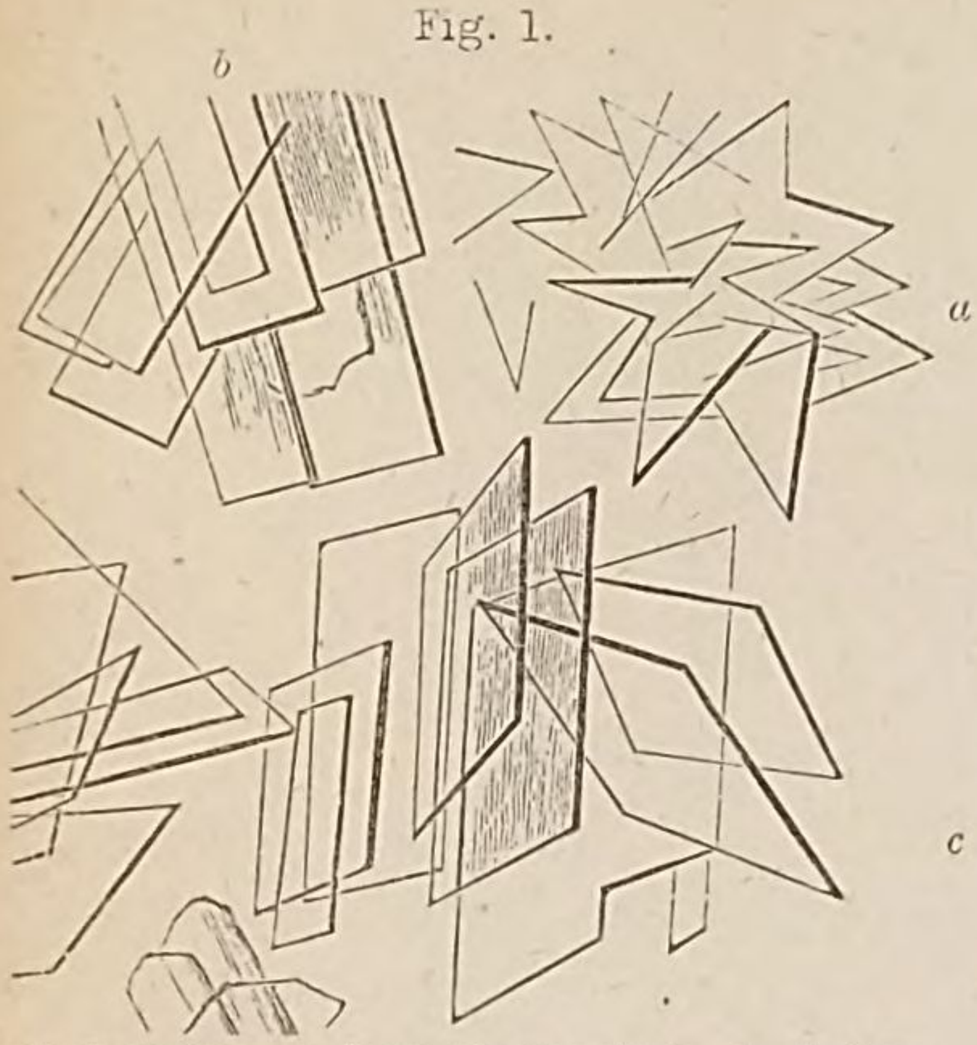
228. Biliary Acids.—Pettenkoffer's Test.—The following directions are given by Pettenkoffer.—Pour a portion of the suspected fluid into a test-tube, and add English sulphuric acid, guttatim, to about $\frac{2}{3}$ the volume of the fluid, whereby the temperature is considerably raised. The addition must be made so gradually that the temperature shall at no time exceed 145° F., as otherwise the choleic acid is too much changed; then add 2 to 5 drops of ordinary cane sugar solution, containing 1 part of sugar to 4—5 parts of water, and shake the whole. If choleic acid be present, a more or less deep violet red colour will be produced according to the amount of bile in solution. Mr. Francis of the Laboratory, Charing Cross Hospital, employs grape sugar instead of cane sugar. A few grains of grape sugar are dissolved in one drachm of concentrated pure sulphuric acid. This he calls the "bile acid test solution." A few drops are allowed to flow into the urine or other fluid supposed to contain bile acids. If these be present

the purple colour will appear just where the two liquids mix together (Dr. Ralfe's "Outlines of Physiological Chemistry," p. 204).

Improved mode of applying Pettenkoffer's Test.—Neukomm (über die Nachweisung der Gallen Säuren, &c., 1860), proposes the following modification. "A single drop of a $\frac{1}{20}$ per cent. solution of choleic or glycocholic acid will yield a splendid purple violet colour if it is brought in contact with a drop of dilute sulphuric acid (4 parts of water to 1 part of sulphuric acid) and a trace of sugar solution, in a porcelain cup and then gently warmed over a spirit lamp. As one cubic centimeter equals about eight drops, it is thus possible to demonstrate $\frac{6}{100}$ milligr. of biliary acid with complete accuracy." As a further test he suggests, "the biliary acid or salt is to be sprinkled with a small quantity of concentrated sulphuric acid, moderately warmed, and then water added. The resinous flocculi that subside are to be separated from the acid, washed with water, but not so as to remove all the sulphuric acid, and then again gently heated in a porcelain cup till coloration ensues. If the residue be taken up in a small quantity of alcohol, and the green solution be evaporated, the interior of the cup will be coated with a deep indigo blue film even when but little acid has been used. If the biliary acids or the sulphuric acid should be impure, or the temperature raised too high, the pigment film will be green." See abstract of Beneke's Memoirs "On the Occurrence, Diffusion, and Action of the Constituents of the Bile in the Animal and Vegetable Organism," by Dr. Duffin, "Archives of Medicine," vol. iv, p. 192, 1865.

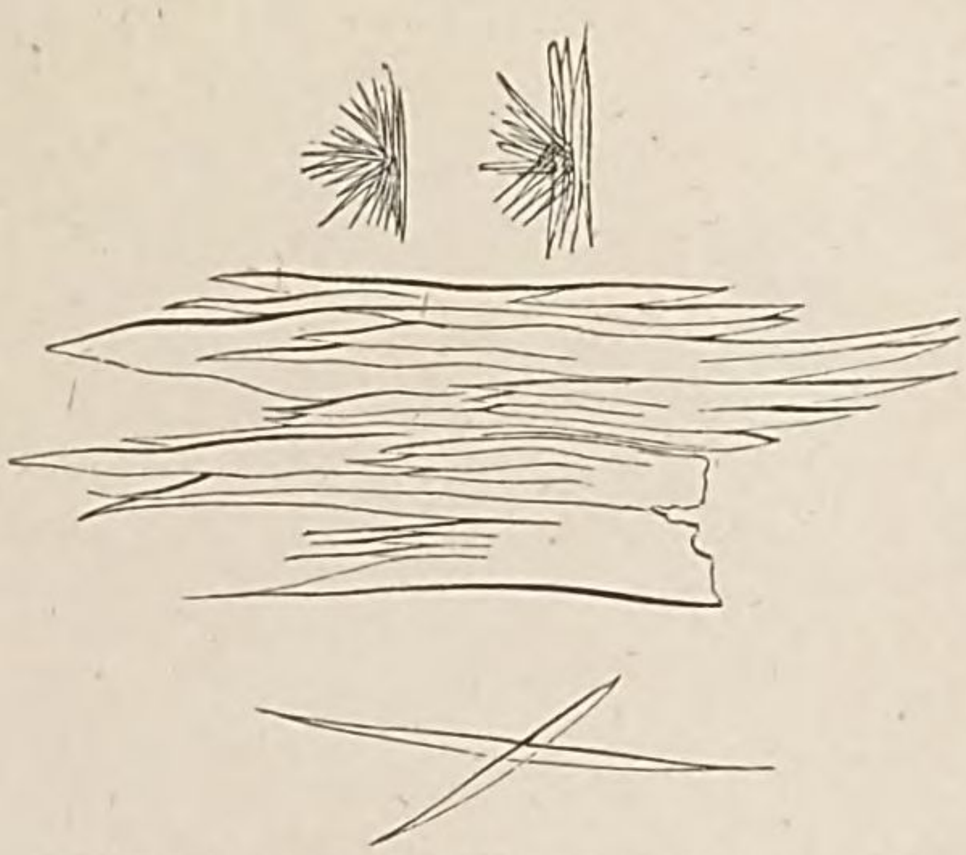
229. Excretine.—This substance was discovered by Dr. Marcet a few years ago. It is present only in human fæces.—"Phil. Trans." 1857, page 403. "Archives of Medicine," No. II, April, 1858. In order to obtain it, the following process is employed. A quantity of excrement is introduced into a long-necked flask, and is exhausted with boiling alcohol ·850. The mixture is filtered, and the solution mixed with a little thick milk of lime, diluted with a quantity of water equal to that of the alcoholic solution. After standing for a few hours, a light precipitate will subside to the bottom of the vessel. This is separated by filtration, washed several times with water, and dried over the water-bath. The dry residue is placed in a flask, and alcohol added. Next a little ether is to be poured in, which much increases the solvent power of the alcohol for excretine. This operation is repeated three or four times the alcohol and ether being allowed to remain on the residue three or four hours before being poured off. The filtered alcoholic solutions are to be evaporated in as cold a place as possible; and after the lapse of a day or two, crystals of excretine will make their appearance (pl. XXI, fig. 7). They are to be separated, and the mother-liquor allowed to remain, that another crop of crystals may form. The impure excretine is to be dissolved in hot alcohol, agitated with animal charcoal, and re-

Fig. 1.



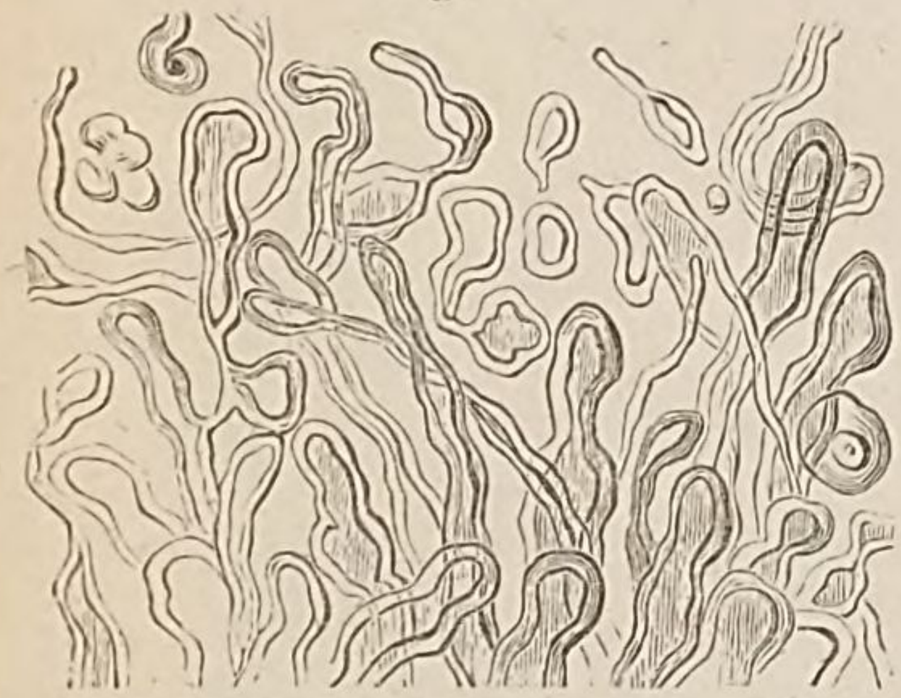
Cholesterine. *a*, from pneumonic lung. *b*, from fluid round an hydatid cyst. *c*, from the brain. × 215. p. 188.

Fig. 2.



Serolin in flakes. After Robin and Verdeil. p. 189.

Fig. 3.



Myelin from the brain × 215. p. 189.

Fig. 7.

Excretine crystallised from an alcoholic solution. × 215. p. 190.

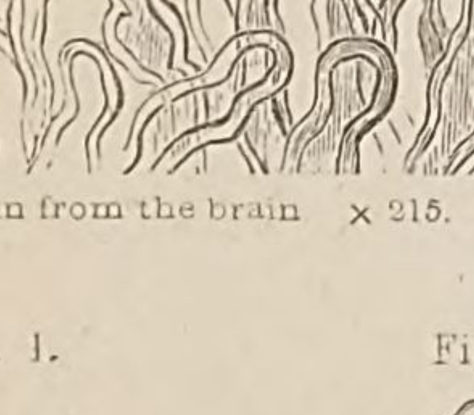
Fig. 1.

Epithelial cell. Air cell of lung Cattle plague. Myelin particles in outer part × 1800. p 189.

Fig. 5.

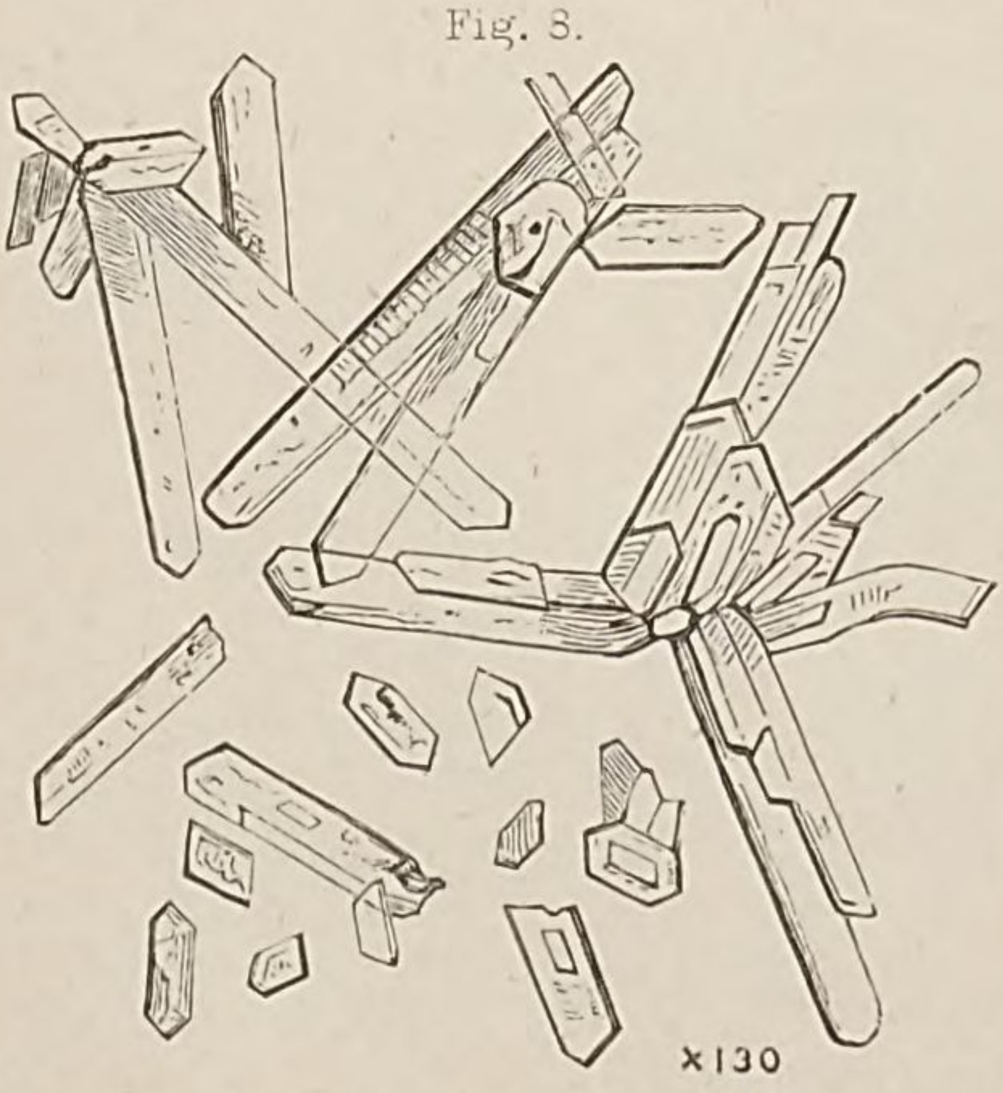
Cerebral matter in water, showing double contours of myelin. × 215, p. 189.

Fig. 6.



Myelin particles from the external portion of cells in air-cells of the lungs. × 2800. p. 189.

Fig. 8.



Crystals of diabetic sugar from urine. p. 196.

[To face page 190.

crystallized. It does not easily crystallize unless its alcoholic solution be allowed to evaporate slowly in a cold place. Frequently nothing more than a few non-crystalline globules are obtained, but these will crystallize if re-dissolved in alcohol and exposed to the cold.

Dr. Marcet has ascertained the composition of excretine to be as follows:

Carbon	80·427
Hydrogen	13·515
Oxygen	3·278
Sulphur	2·780

Its atomic weight is 578, assuming that an equivalent contains one equivalent of sulphur.

230. Crystallizable Substances from the Blood.—The beautiful compound known as *Hæmato-globulin*, *Hæmoglobin Hæmato-crystallin* is held in solution in the red blood-corpuscles of man and animals. It was examined first by Funke. Soon afterwards Kunde published important memoirs on the subject.* Blood crystallization has been carefully investigated by Lehmann.† In the "Medical Times and Gazette" for 1852, will be found a very interesting paper on the subject by Dr. Parkes.‡

The crystals are very readily obtained, for microscopical examination from some kinds of blood, by diluting it with some fluid. Some of the best examples of blood crystals are figured in pl. XXII, page 194. A drop of fresh blood may be placed upon a glass slide, and after the addition of a drop of water, alcohol, or ether, the whole should be lightly covered with thin glass. A hair, or a small piece of thin paper or wood, may be placed between the glasses, in order that a stratum of fluid of sufficient thickness may be retained. It is preferable to use defibrinated blood. Often the corpuscles and a little serum may be removed from the clot by firm pressure, and from this, very perfect crystals may frequently be obtained. The blood-corpuscles do not, as is still repeatedly stated to be the case, become *ruptured* by endosmosis, their contents being set free and undergoing crystallization as the solution gradually becomes concentrated by spontaneous evaporation which goes on at the edges, but the soft semi-fluid matter of which the entire red blood corpuscle is composed, passes from its colloid to its crystalline condition. Although under many circumstances the outer part of the corpuscle may become hardened so as to give rise to a firm envelope,

* Dissert. inaug. Lips. 1851 (O. Funke). Zeitschrift f. rat. Med. N. F. Bd. I, II. See also Funke's "Atlas of Plates," translated by the Cavendish Society, 1853.

† "Lehrbuch d. Physiolog. Chemie," vol. i, second edition, 1853. Bei. d. k. Sächs. Gesel. d. Wiss., 1852—1853.

‡ See also a paper on "Albuminous Crystallization," by Dr. Sieveking, in the "British and Foreign Medico-Chirurgical Review," for October, 1853, in which some excellent woodcuts of blood-crystals are given. Dr. Hoppe Seyler's "Handbuch der Physiologisch-und Pathologisch-Chemischen Analyze" should also be consulted.

this is not a necessary structure, and there is no true cell wall developed upon the surface of the red blood-corpuscle. See Chapter XIII. The time which elapses before crystallization takes place varies from one hour to several hours, or days, in specimens of blood from different animals. The blood-corpuscles of the Guinea-pig are most favourable for experiment, as the matter of which they are composed crystallizes very readily. Sometimes the blood-corpuscles pass into the crystalline state in the course of twenty minutes.

The form of the crystal often varies slightly in the same specimen, but the blood of different animals yields crystals of very different forms. The prismatic form is that most commonly obtained from the blood of man, the carnivora, and fishes. Tetrahedral crystals are met with in some of the rodentia, as the Guinea-pig, pl. XXII, fig. 1, while six-sided tables are formed in the blood of the squirrel, mouse, and some others, fig. 6. Professor Lehmann obtained beautiful rhomboidal crystals from the blood of the hamster (another of the rodentia). Blood crystals form more readily in daylight than in the dark, but most rapidly when the slide is exposed to the light of the sun. I have not succeeded in obtaining crystals of the blood of the ox or sheep. From pig's blood crystals were obtained with some difficulty, after passing oxygen and carbonic acid through the fluid and diluting it with alcohol and water. The crystals from pig's blood are in the form of prisms and acicular crystals. Frog's blood cannot readily be made to crystallize: Teichmann has however succeeded in obtaining crystals from frog's blood, by the addition of a large quantity of water at a very low temperature. Zeitschrift für rat. Med. N. F. Band III, Heft 3.—"British and Foreign Medico-Chirurgical Review," April, 1854. Professor Lehmann has obtained crystals readily from the blood of the Italian lizard.

I have often obtained crystals from Guinea-pig's blood without the addition of any fluid, and without any evaporation whatever. One blood corpuscle becomes a single crystal, and if the slide on which the drop of blood is placed and covered with thin glass be gently warmed, the corpuscles break up, and each little fragment assumes the crystalline form, and becomes a minute but perfectly formed tetrahedral crystal. This is a very important fact in connection with the question of the constitution of the red blood corpuscle. See Chap. XIII. Dog's blood crystallizes in the course of a short time upon the addition of a little alcohol. Human blood crystallizes after the addition of water, slowly, if only just removed from the body, but more rapidly if the blood be not quite fresh. The crystals shown in pl. XXII, figs. 2, 4, were obtained by diluting a drop of fresh blood from the finger, with a drop of distilled water; and after covering the mixture with thin glass, the slide was placed in a light place. Crystallization commenced about forty hours after the addition of water to this specimen of blood.

Lehmann discovered a process by which large quantities of blood crystals might be prepared. This consists in passing oxygen and carbonic acid through the blood which has been diluted with much water. The blood which answers best, is that of the dog and Guinea-pig, but as far as I know, no one has obtained crystals from the blood of the ox or sheep. This depends probably upon the material of the blood-corpuscles being less readily dissolved than that of most animals. The following plan yielded an abundant quantity of crystals from the blood of the dog and Guinea-pig. The defibrinated blood was diluted with half its volume, or with an equal volume of water. Sometimes it was necessary, in the case of dog's blood, to add a little alcohol or ether until the solution of the corpuscles had taken place, which can always be ascertained by microscopical examination. Through the solution, oxygen was passed for a quarter or half an hour, and then carbonic acid was transmitted through the same fluid during half the time the oxygen had been passed. In the course of an hour, or longer, an abundant precipitate, consisting entirely of blood crystals, was produced. This was separated by filtration, and dried. If the crystals are required quite pure, they must be re-dissolved in water until the mixture has a specific gravity of between 1004 and 1002, and then alcohol must be added until the specific gravity is reduced to ·970. Crystals will be deposited in a few hours.

Dr. Teichmann pursues rather a different plan for obtaining blood crystals. After separating the serum and fibrin as far as possible, the blood is diluted with four or five times its bulk of water. The fluid is precipitated with sulphate of copper. The precipitate is washed and pressed well, but not dried. It is extracted with alcohol containing about one part of concentrated sulphuric acid, to three hundred parts of alcohol.* Another plan, the details of which were perfected by Hoppe Seyler, is to allow blood to flow into a vessel cooled by ice. The fibrin is to be removed in the usual manner, and a 10 per cent. solution of common salt added. When the red corpuscles have settled, the supernatant fluid is poured off, and the sediment thrown upon a filter and well washed with the salt solution. The sediment is then to be treated with a mixture consisting of four volumes of ether and one of water. The *red solution* which results is to be filtered into a vessel placed in ice. Alcohol is then poured in until a slight turbidity appears. The whole is then left for crystallization to take place. Extreme cold is required if it is desired to obtain the hæmato-crystallin of the blood of some animals in its crystalline form. Although the chief constituent of the red blood corpuscles of some animals has not yet been obtained in its crystalline form, there is good reason for concluding

* Henle and Pfeuffer's "Zeitschrift," vol. viii, page 141.

that every kind of hæmato-crystallin is capable of crystallization. The spectroscopic examination of the crystallizable substance of the blood is considered in Chapter XI.

I have found great difficulty in preserving many specimens of blood crystals as permanent objects. I have succeeded, however, in keeping some human blood crystals mounted in the dry way; crystal from dog's blood have been mounted in Canada balsam, while the beautiful octahedral crystals from Guinea-pig's blood have kept pretty well in the fluid to which spirit had been added, but these soon lost their brilliancy, and changed to a dull greenish brown colour. In glycerine, crystals from Guinea-pig's blood have been preserved for many years. Crystals of hæmatoidin may be readily preserved in glycerine or in Canada balsam.

Hæmatin may be obtained from hæmato-crystallin. It occurs in old extravasations of blood, and may be detected in the fæces. It forms crystals of a very dark colour, which are insoluble in water, alcohol, and ether. It is found amorphous. When dried it forms a brown powder, containing nearly 9 per cent. of iron. A thin layer of a solution of hæmatin appears of a greenish colour, while a thick one is dark red. Solutions of this substance differ in colour according as they are examined by transmitted or reflected light. Hoppe Seyler names a substance closely allied to hæmatin, methämoglobulin. This may be a mixture of hæmatin and some albuminous substance.*

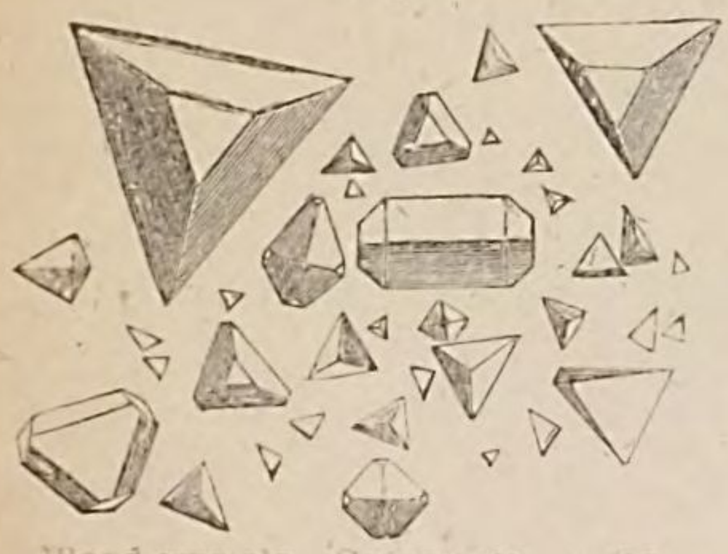
Hæmatoidin is a modified form of hæmatin. It is not easily decomposed, is insoluble in water, alcohol, ether, and acetic acid, but readily soluble in alkalies. This is the substance which is found in old clots and extravasations, and not unfrequently in the walls of some of the smaller vessels, perhaps marking the situation of old hæmorrhages. It crystallises in very beautifully defined rhombic crystals, pl. XVII, fig. 3. It also forms long filaments, and not unfrequently slightly curved elongated crystals, collected into bundles, which sometimes take the form of oval and dumb-bell shaped masses, figs. 8, 9. This substance seems closely allied to a yellow crystalline material obtained from bile. It would indeed be very difficult to distinguish hæmatoidin crystals found in clots from some crystals which have been found under certain circumstances in biliary matters. Hæmatoidin may therefore be the

* The following references to papers on blood crystallization will be useful to those who wish to perform original investigations :—

Nasse in "Müller's Archiv.," page 439. Kölliker in "Zeitschr. f. Wiss. Zool.," 1849, i, page 260. Reichert in "Müller's Archiv.," 1849, page 197. Remak in "Müller's Archiv.," 1851, page 481. Funke "Zeitschr. f. rat. Med. N.F.," i, pages 184, 192; ii, pages 192, 288. Kunde "Zeitschrift f. rat. Med. N.F.," ii, page 271. Lehmann in "Ber. der k. Sächs. Ges. d. Wiss. zu Leipzig," 1852, pages 23, 78; 1853, page 102. Robin and Verdeil, "Traité de Chimie Anatomique et Physiologique," ii, page 335. Teichmann, in "Zeitschr. f. rat. Med. N.F.," iii, p. 375, viii, page 141.

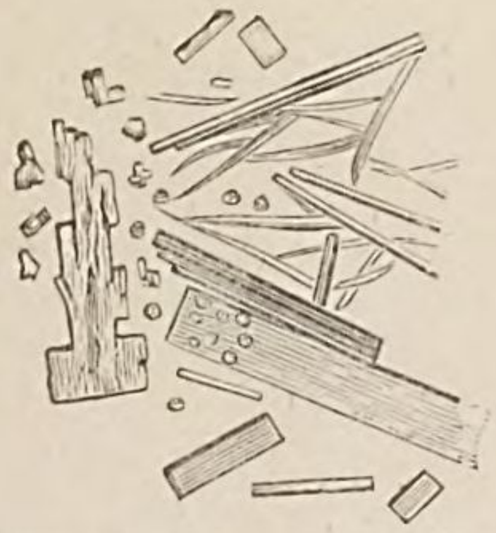
BLOOD CRYSTALS.

Fig. 1.



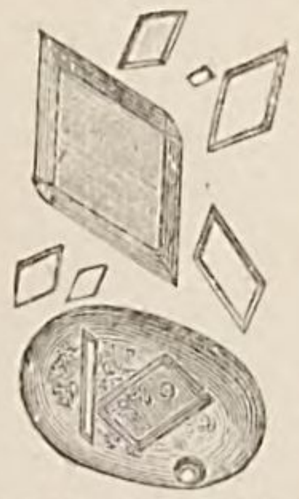
Blood crystals. Guinea-pig. × 215 p. 192.

Fig. 2.



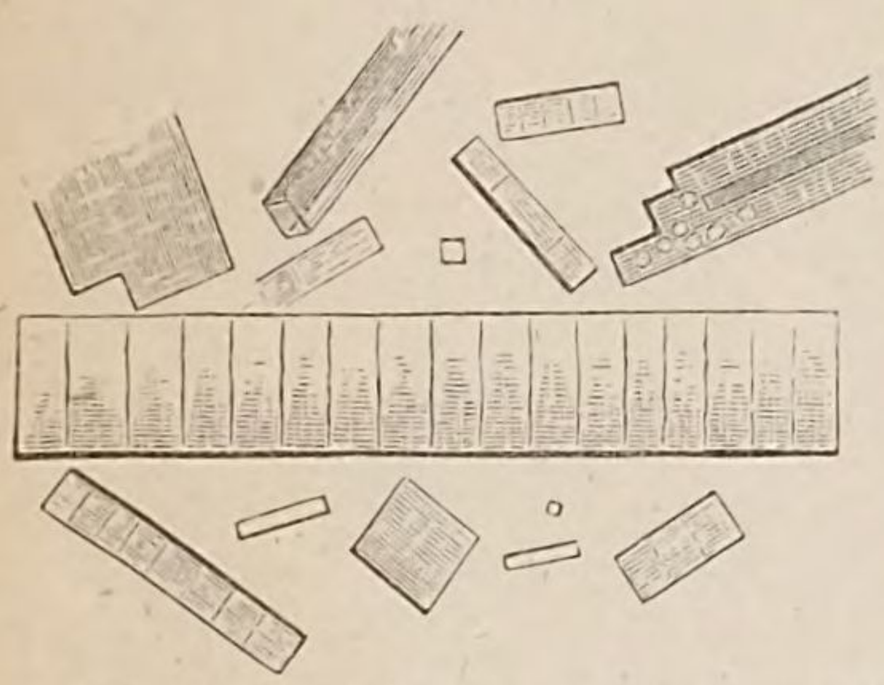
Human blood crystals. × 215. p. 192.

Fig. 3.



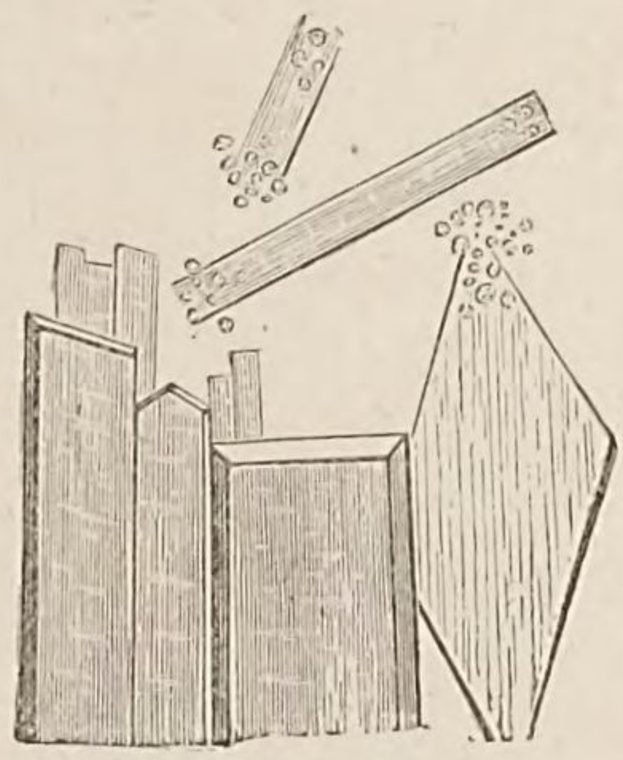
Crystals of hæmatoidin from human liver × 215. p. 193.

Fig. 4.



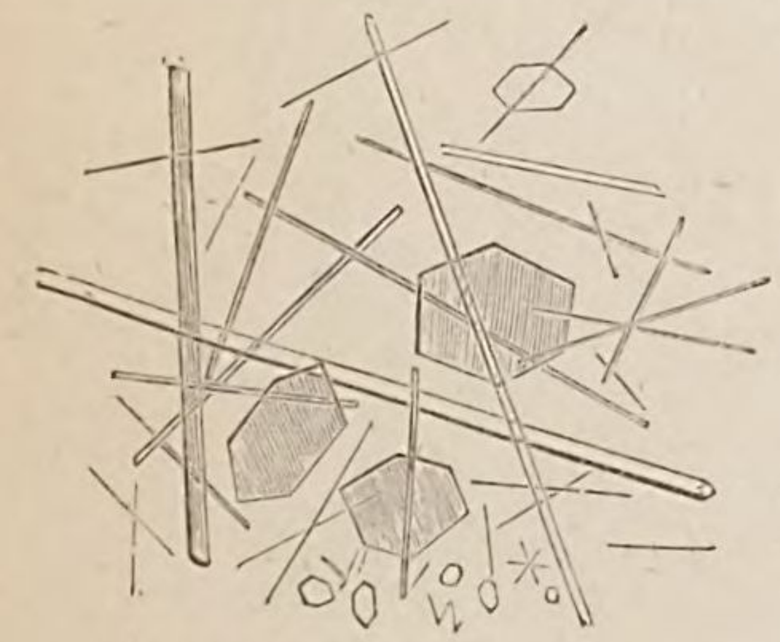
Blood crystals. Human. × 215. p. 192.

Fig. 5.



Blood crystals. Cat. × 215.

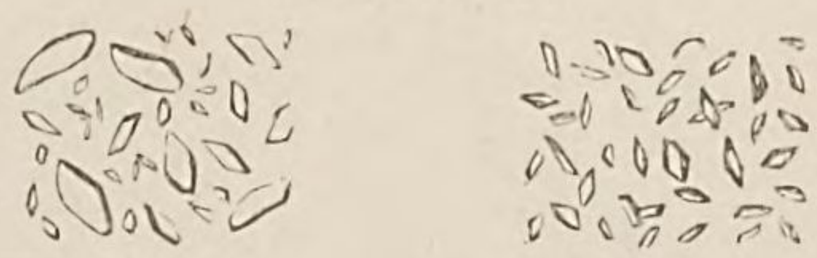
Fig. 6.



Blood crystals. Mouse.

Fig. 7.

HÆMIN CRYSTALS



Human. × 215

Pig. × 215.

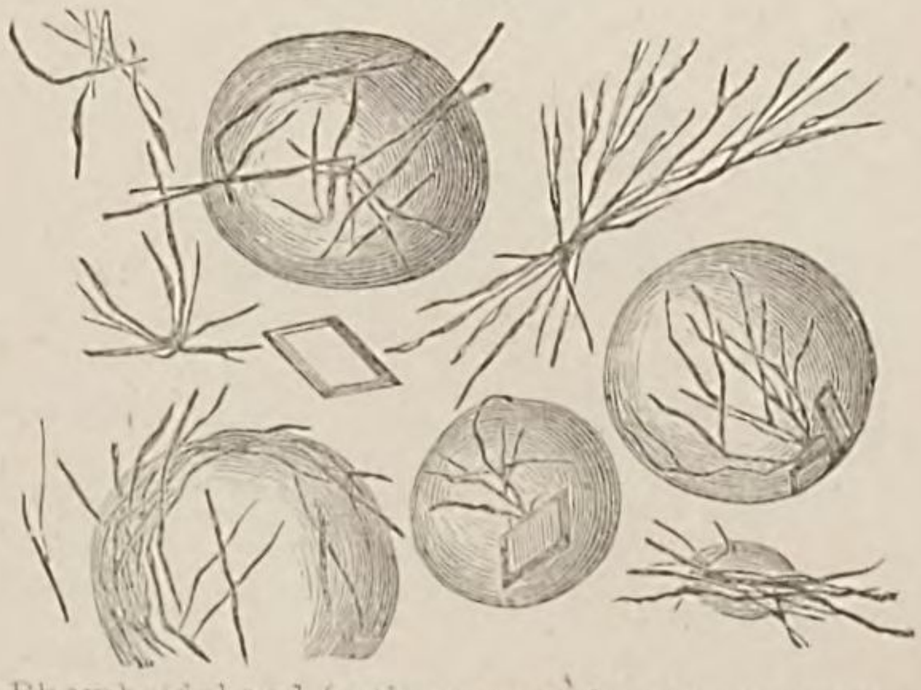
Toad. × 215. p. 195

Goldfinch. × 215

Fig. 8.

Feathery crystals of hæmatoidin, found in the urine a fortnight after slight rupture (?) of one kidney. Human subject. × 215. p. 194.

Fig. 9.



Rhomboidal and feathery crystals of hæmatoidin from a softened clot. Human. × 215. p. 194.

$\frac{1}{1000}$ of an inch |——| × 215.

[To face page 194.

same substance as that which has been obtained from bile under the names *Cholepyrrhin*, *Biliphæin*, *Bilifulvin*, and more recently, *Bilirubin*. Zenker and Funke have indeed shown that from the yellow crystals of Bilifulvin, red crystals of Hæmatoidin may actually be obtained.

The tests for the colouring-matters and other constituents of the blood will be referred to in Chapters XII and XIII.

Hæmin. Dr. Teichmann also obtains beautiful crystals of a dark red colour, by treating the clot of blood, moist or dry, with glacial acetic acid. He tells me that the crystals of hæmin thus obtained have the same form in all animals, while the crystals just described differ much in form and colour in different animals. A small quantity of common salt may be added to the blood, upon a glass slide, next a drop or two drops of glacial acetic acid, and the slide gently warmed. The crystals of hæmin soon separate. Blood that has been kept for some time yields these crystals as well as fresh blood. Crystals of hæmin obtained from the blood of the human subject, pig, toad, and goldfinch are represented in pl. XXII, fig. 7.

231. Crystallization of Bile.—The glycocholates of potash and soda were first obtained in a crystalline form by Platner. The crystallizable substance of the bile may be readily obtained as follows :—Perfectly fresh ox-bile is to be rapidly evaporated to dryness over the water-bath, and the dry residue powdered and extracted with absolute alcohol ; the dark green alcoholic solution is then quickly filtered into a small flask or bottle, and ether gradually added until the white precipitate at first formed ceases to be re-dissolved upon agitation. Care should be taken to add the ether *very gradually*, or a bulky amorphous precipitate may occur, which will not afterwards crystallize. The bottle is to be lightly corked, and allowed to stand in a still place. After a few days, stellar masses of beautiful and almost colourless crystals appear ; these increase until tufts of a considerable size are produced. The crystals may be subjected to microscopical examination, immersed in a drop of the solution in which they were formed ; or they may be carefully washed with alcohol, to which a tenth of its bulk of ether has been added, and rapidly dried in vacuo. Crystallized bile is represented in pl. XXIII, fig. 1. An excellent paper "On the Constitution and Physiology of the Bile," by Dr. Jno. C. Dalton, junr., will be found in the American "Journal of the Medical Sciences," translated by the Cavendish Society, for October, 1857. *See* also "Lehmann's Chemistry."

When dried, the bile crystals may be mounted in a cell from which the air is completely excluded. If exposed to the air while moist, they rapidly deliquesce. I have preserved some of these crystals, in the solution in which they were formed, in a thin glass cell for some months. Ox-bile and pig's-bile may be crystallized, but no one has yet succeeded in obtaining crystals from human bile. Sometimes considerable difficulty

is experienced in causing bile to crystallize, and it may be necessary to make repeated attempts with perfectly pure alcohol and ether before the result is satisfactory.

Crystals of glycocholic acid, of taurocholate of soda and of cholic acid are represented in pl. XXIII, figs. 2, 3, 4.

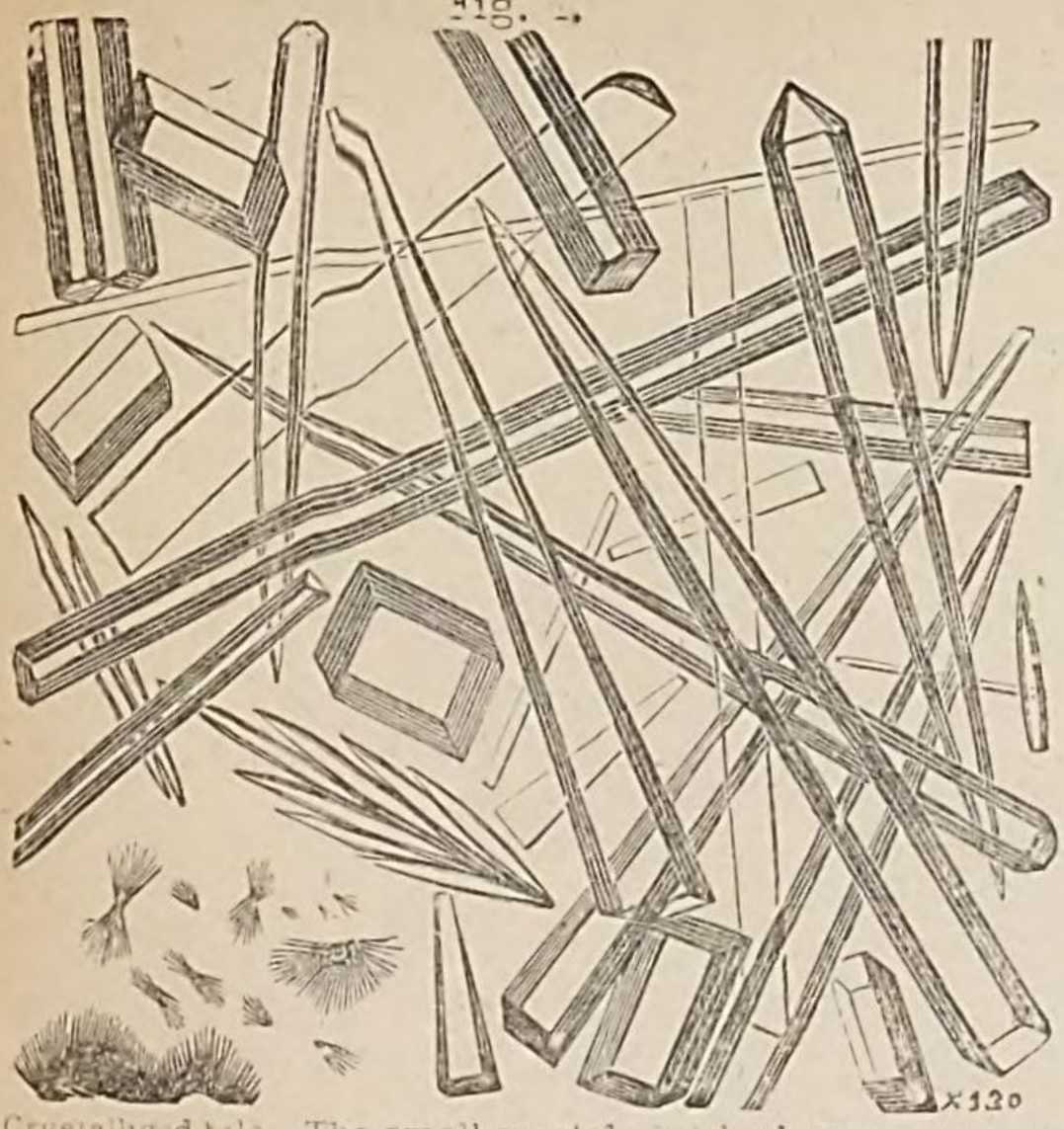
232. Animal Sugar.—Among the crystalline substances to be obtained from animal substances must be included a form of glucose or sugar nearly allied to, but not identical with, the sugar of fruits. Of animal sugars there are several varieties, some of which crystallise very readily, while many have not yet been obtained in a crystalline form. Diabetic sugar oftentimes crystallises so easily that if a drop of urine, which contains a large quantity, be allowed to evaporate spontaneously in a warm place on a glass slide beautiful well-formed crystals will be slowly formed. On the other hand, the sugar obtained from the liver does not readily crystallise. I have figured crystals of animal sugar from diabetic urine in plate XXI. *See* also "Kidney Diseases, Urinary Deposits, and Calculous Disorders," 3rd edition.

Animal sugar is formed in all animals from an amyloid matter which is produced in considerable quantity with other substances, which together constitute the outer part of the so-called "liver cell." During the disintegration of this formed material, biliary matters, fats, and other chemical products, are at the same time developed. This *amyloid*, *glycogen*, or *glycogenic matter* is allied to starchy and saccharine substances, but it is not identical with any vegetable amyloid. It was discovered by Bernard in the liver in 1857.

Glycogen is amorphous, tasteless, and without colour. It is soluble in water, from which solution it may be precipitated by alcohol or ether. Saliva, pancreatic secretion, and many animal matters, especially if in a state approaching decomposition, cause its conversion into dextrine and grape sugar. The same change is effected by boiling with dilute hydrochloric or sulphuric acid. Glycogen is always present in the perfectly fresh liver of all animals, and may be easily extracted from the hepatic tissue by water, the albumen taken up with it being coagulated and separated by filtration.

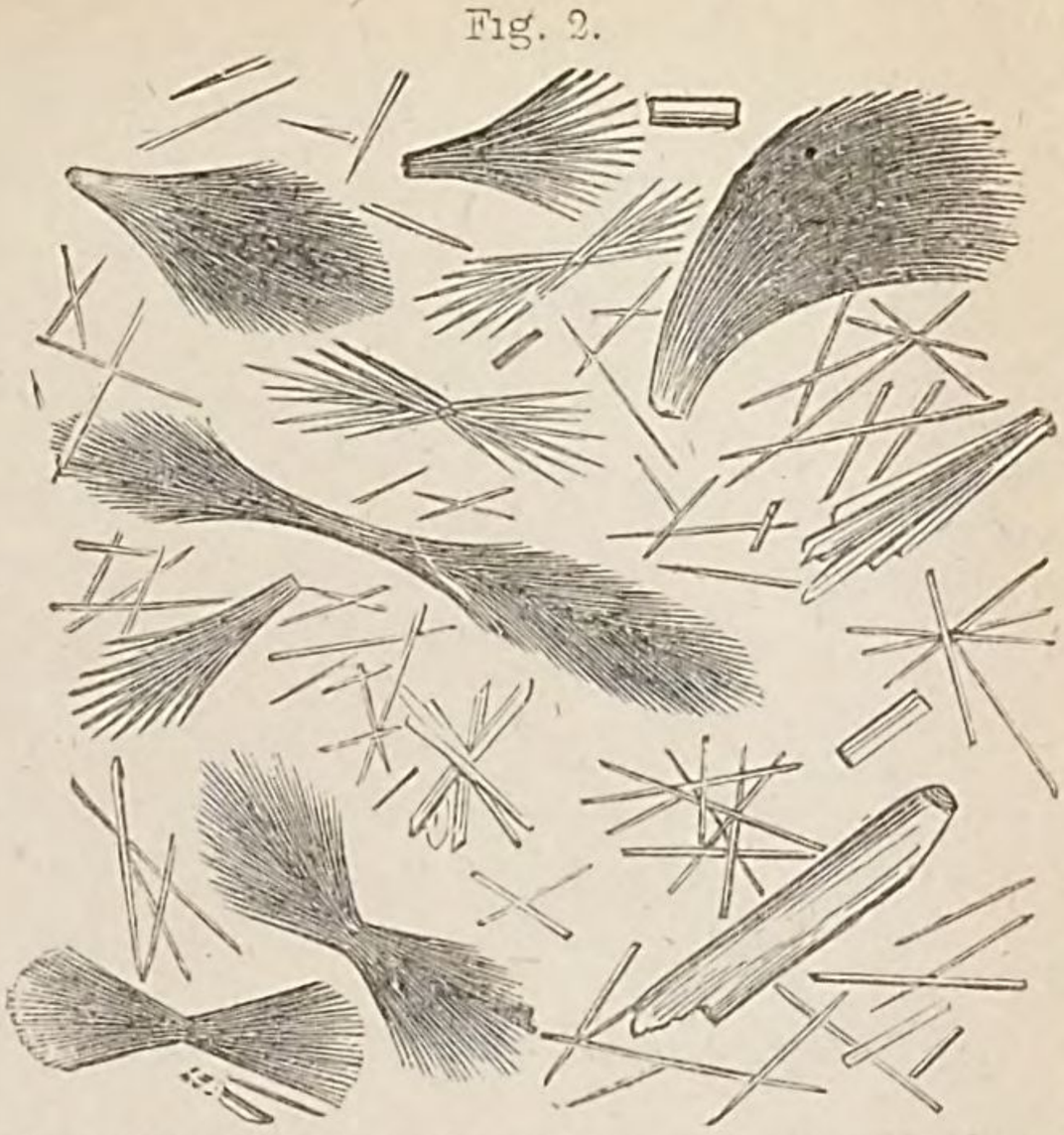
Within a few hours after death the glycogen, in contact with animal matter, undergoes conversion. If, then, the liver infusion be examined some time after death sugar will be found in large quantity. It has been, however, proved experimentally that sugar may be detected in the liver within a few seconds after death, and I think there is little room for doubting that animal sugar is *formed* during life. Sugar is a very soluble and rapidly diffusible substance. It is therefore quickly removed. It is indeed carried off by the blood and changed as fast as it is formed from the glycogen produced as a consequence of vital changes in the living animal. After death, and in certain forms of disease, sugar is

Fig. 1.



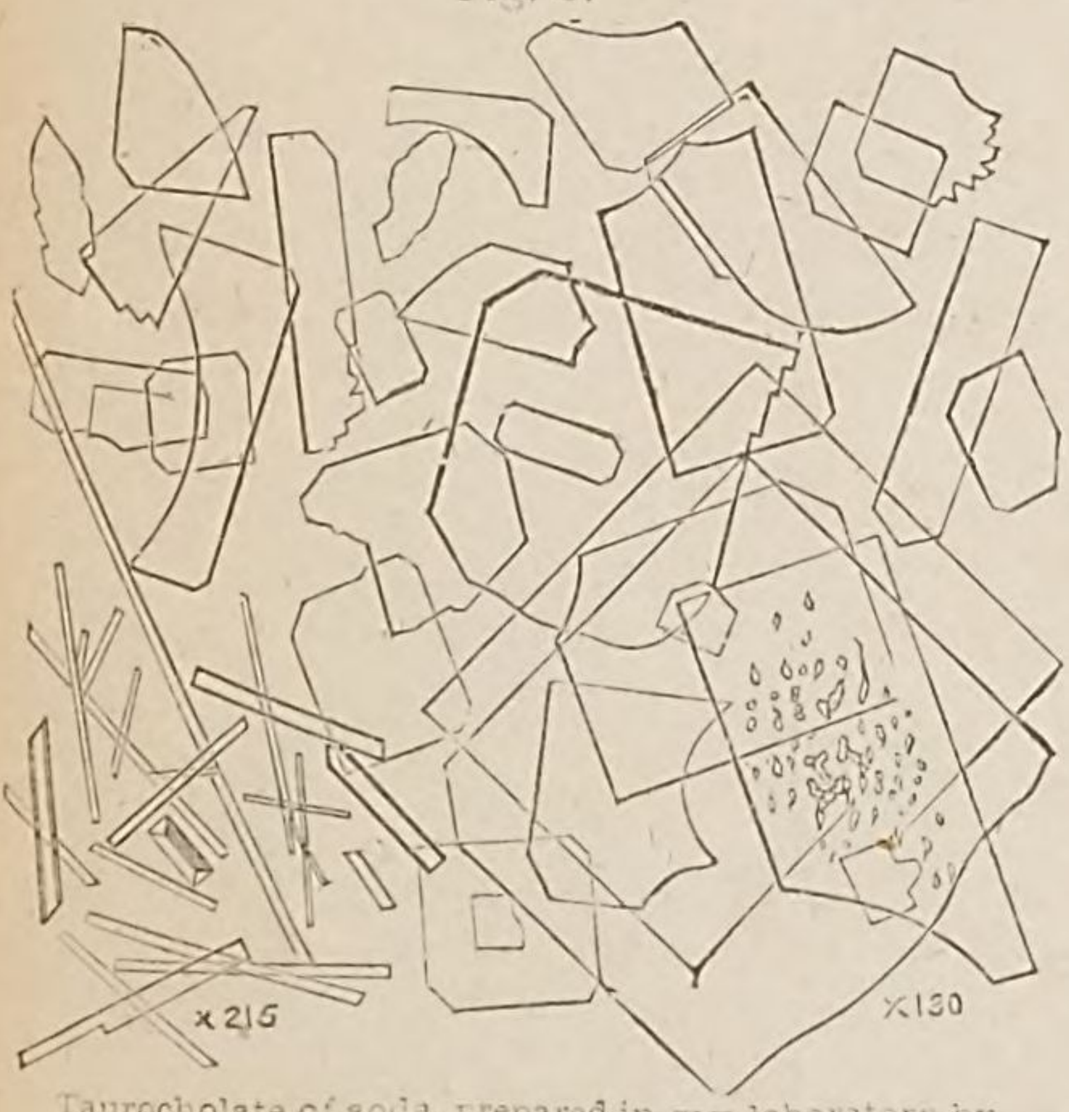
Crystallised bile. The small crystals in the lower corner on the left are drawn as they appeared to the unaided eye. p. 196.

Fig. 2.



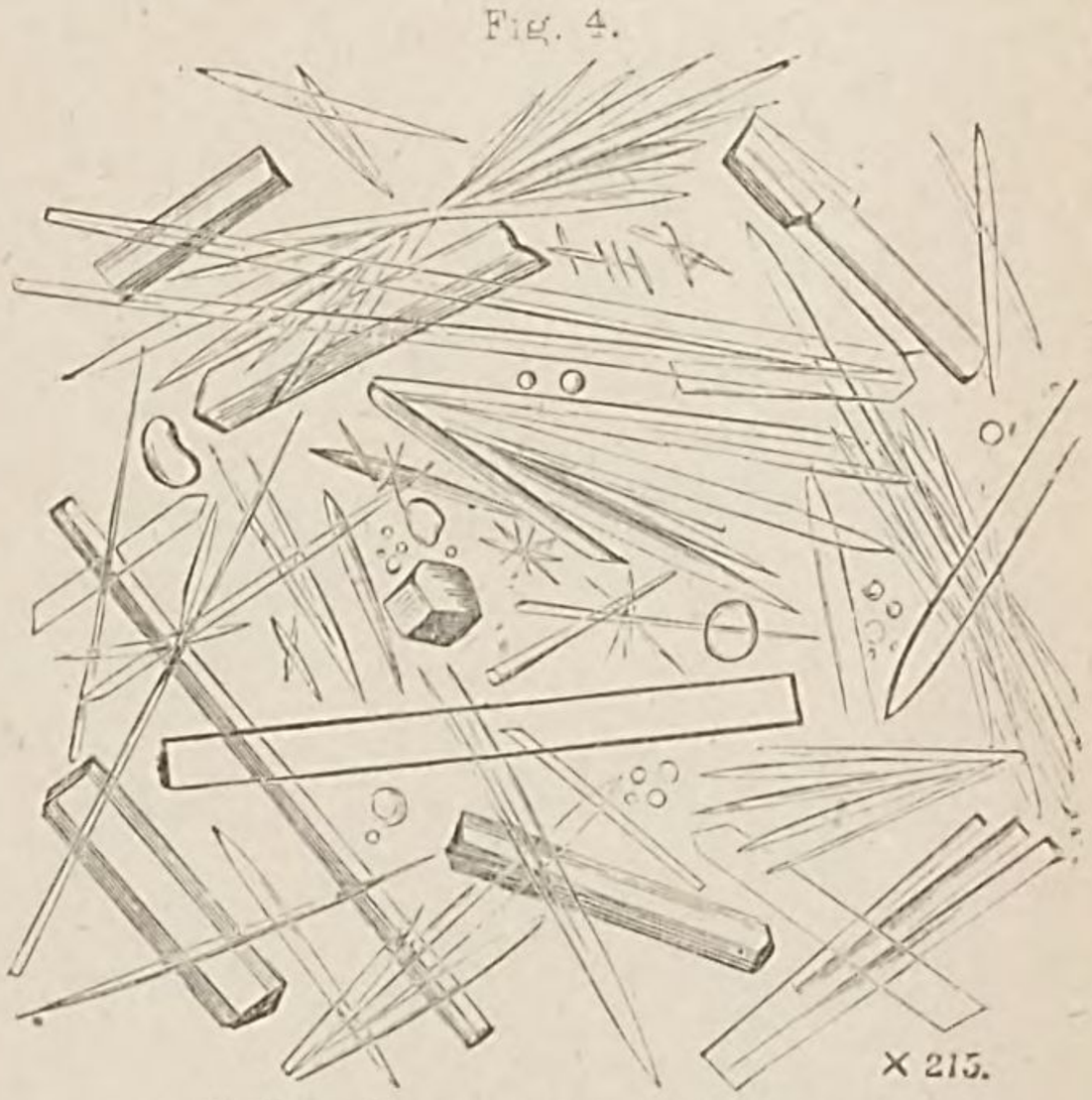
Glycocholic acid, prepared by decomposing glycocholate of lead with sulphuretted hydrogen. p. 196.

Fig. 3.



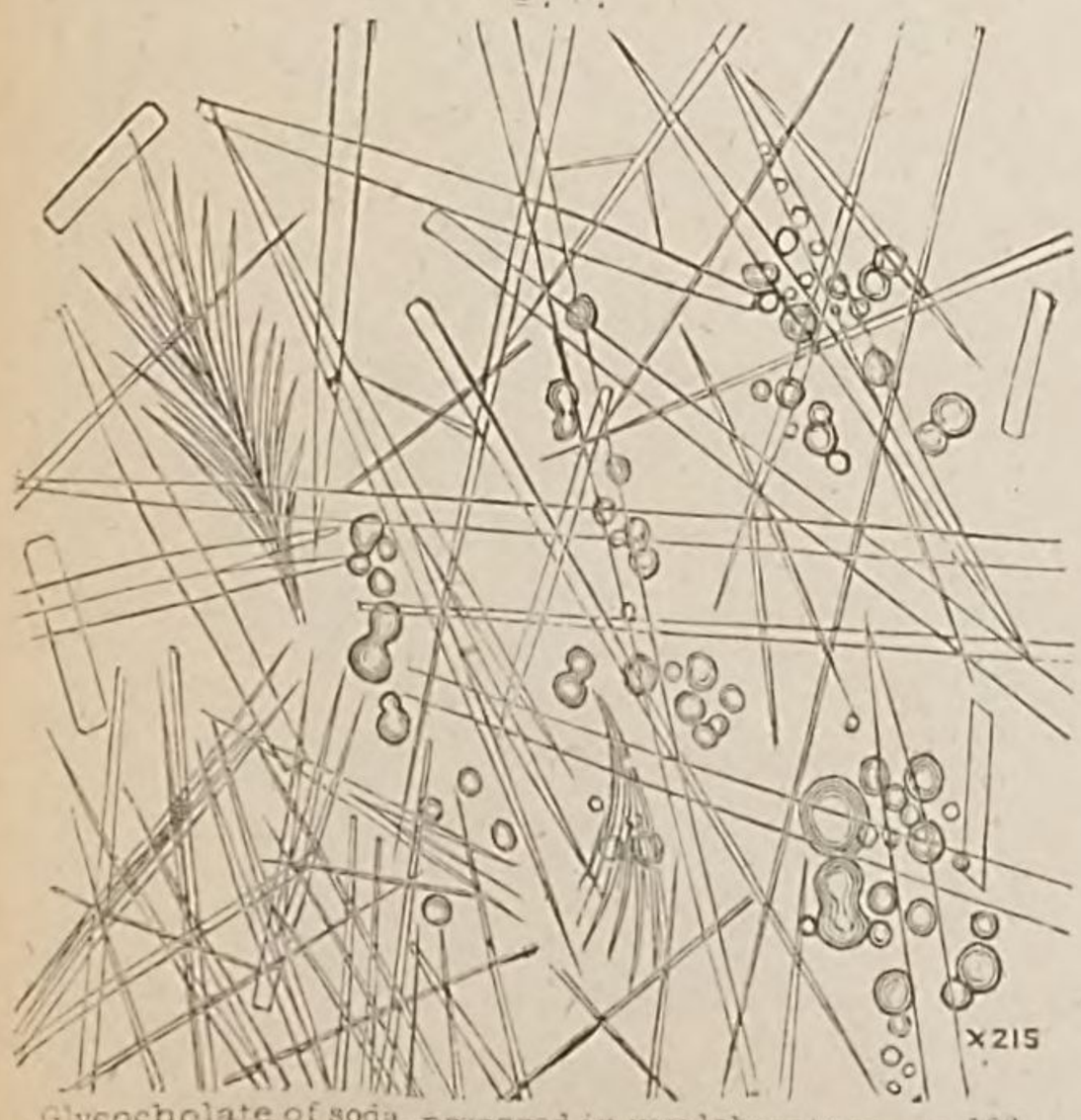
Taurocholate of soda, prepared in my laboratory by Dr Von Bose.

Fig. 4.



Cholic acid, prepared in my laboratory.

Fig. 5.



Glycocholate of soda, prepared in my laboratory p. 196.

Fig. 6.

Crystals of chloride of ammonium, obtained by breathing for several minutes upon a glass slide, moistened with hydrochloric acid. p. 147.

1000th of an inch |___| × 130.

" " |___| × 215.

[To face page 196.

not destroyed as in the normal condition, but it accumulates in enormous quantity, and in certain conditions is no doubt produced much faster than it can be decomposed and got rid of. In such cases sugar may be detected in the blood and in all the fluids and tissues of the body.

Much unprofitable discussion has been carried on concerning the origin of the glycogen. It has been very safely asserted that it is derived from the food—as if there were constituents of the body derived from other sources. Every tissue and every constituent of the organism, it may be said, is derived from the food, but glycogen and every substance peculiar to the organism is made in the body out of living matter peculiar to the living organism. These things cannot be made out of the body from the food by any means yet discovered. Glycogen is produced in animals living upon purely animal or purely vegetable matter. Its formation would seem to depend far more upon the peculiar formative powers of the bioplasm of the liver cell than upon the nature of the food introduced. Glycogen is, in fact, formed from the living matter of this cell, and does not result from changes of a merely chemical nature in certain constituents of the food, as has been supposed by some. But glycogen is found in many tissues. It is to be detected in the textures of young animals generally, and may be extracted from the placenta. It exists in the juice expressed from muscular tissue, and is probably present in smaller proportion in many other textures, and in some fluids. Glycogen is precipitated from its aqueous solution by alcohol.

233. Removing Stains from the Hands.—In many chemico-microscopical investigations it is impossible to avoid staining the fingers, and it may be well to give a few hints in this place as to the removal from the cuticle of different varnishes and other materials used by the microscopist. *Brunswick black* may be removed with turpentine or by rubbing on lard or oil, which may afterwards be washed off with soap and water. *Marine glue* can be peeled off, or it may be dissolved by a little ether, chloroform, or coal naphtha. *Canada balsam* by turpentine, ether, or chloroform.

Chromate of lead and other chromates stain the cuticle of a deep yellow colour to be removed by hydrochloric acid and subsequent washing. The stains from *Prussian blue fluid* may be got rid of by dilute potash and often by ordinary washing alone. *Carmine stains* are obliterated by first washing the reddened cuticle with ammonia and then with hydrochloric acid. *Sealing-wax* and other spirit varnishes may be removed by washing the cuticle with alcohol.

CHAPTER XI.

Spectrum Microscopic Analysis.—Spectrum Microscope.—Of examining Substances by the Spectrum Microscope.—Crystals.—Blowpipe beads.—Examination of Solutions.—General outline of Comparative Chromatology.—Spectrum-Microscope applied to Physiological Inquiries.—Improvements in the Spectrum.—Method of detecting blood, by H. C. Sorby, F.R.S.

Of all the methods for detecting chemical substances in a solid, liquid, or gaseous state, that with the aid of the spectroscope is the most delicate. Sir David Brewster claims to have been the first to have employed spectrum analysis, but the process was first brought to perfection by Bunsen and Kirchoff of Heidelberg, by whose wonderful discoveries entirely new fields of research have been opened. Improvements in the method of observation have since been made by a great number of observers, and the spectroscope has lately become a valuable instrument of chemical research.

This method of analysis has been recently applied by Sorby to the microscope. Already many important steps have been gained, and it is probable that much will be discovered by this new method of inquiry. The chief value of the spectrum microscope is that it enables us to determine with care and certainty, and in a very short space of time, the nature of many organic substances of very complex composition, for a great number of which no other means of detection and demonstration are known.

234. Spectrum Microscope.—The spectroscope was first adapted to the microscope by Mr. Sorby ("Quarterly Journal of Science," 1865), who employed for obtaining the spectrum a simple triangular prism placed below the achromatic condenser. This plan was, however, afterwards much improved by Mr. Sorby and Mr. Browning, and the prism was placed *above the upper glass of the eye-piece.* The structure of the prism itself will be understood by reference to pl. XXIV, fig. 1. It consists of "two rectangular prisms of flint glass, between which is a rectangular prism of crown glass, and at each end another prism of crown with an angle of about 75°." These are all connected together with Canada balsam. The compound prism thus prepared is placed between two pieces of blackened cork, and inserted into a tube having a cap with an elongated opening at *a*, and a circular stop at *b*. The lower part of the tube fits on to the eye-piece as shown in the fig. 1.

The beam of light admitted must be narrow, or the dark lines produced will not be distinct. It is desirable that there should be means for reducing or increasing the width of the narrow slit, and this is effected in Mr. Sorby's instrument by the aid of a screw. The arrangement will be understood by reference to fig. 3. The screw by which the slit may be altered in breadth is marked by a^* in figs. 1 and 3. The upper lens of the eye-piece should be achromatic, and Mr. Sorby and Mr. Browning have arranged a special eye-piece for spectrum observations, the general structure of which will be understood by reference to the figure.

In using this spectrum eye-piece, the object to be examined should be selected with the ordinary eye-piece of the microscope, which should then be removed and the other placed in its stead, but, as Mr. Sorby has remarked, it is practically more convenient to use a *binocular*, as the spectrum eye-piece can be adapted to one tube, while the other is provided with the ordinary eye-piece; by the latter, the object can be carefully selected and immediately afterwards examined by the former, without its position being disturbed.

In order that the whole spectrum should be in focus at the same time, Mr. Sorby has had the upper lens (eye-glass) of the eye-piece rendered achromatic; and, for the convenience of comparing two spectra side by side, he has adapted to the side of his special eye-piece a stage with a prism so arranged as to reflect the rays towards the eye-piece. The achromatic lens is represented at *c*, fig. 1, the slit is seen at *d*; *e* is a right angled prism which extends half over the slit, and the light transmitted through this from the side stage *f*, passes through the slit a little on one side of the centre, while the light which comes through the body of the microscope, passes through the slit a little on the other side of the median line, as shown by the lines *g h*. When the analysing prism, *i*, is placed over the eye-piece, two spectra will therefore be seen, which are situated close together, and can be easily compared. The two spectra may readily be made of equal brightness by regulating the width of the opening in the stage attached to the eye-piece.

By varying the width of the slit, it is easy to modify the spectrum in such a manner as to see the absorption bands to the greatest advantage, but there are limits beyond which this should not be done; and we must then alter the thickness of the object, or the strength of the solution, if it is dissolved.

235. Of examining Substances by the Spectrum Microscope, and of the "Absorption Bands" seen.—There are many substances which although transparent, completely obstruct the rays passing through certain parts of the spectrum, thus producing dark lines technically called "absorption bands." Different substances give rise to "bands" in different parts of the spectrum; sometimes a portion of the red light is cut off, sometimes a portion of the green, and so on. The bands are

sometimes very numerous. In some cases they are broad and comparatively faint, and in others they are very narrow, sharp, and well defined. The number and position of these bands may be very different, even though the general colour of the substances may be nearly the same, and thus we are enabled to distinguish them from one another.

It would, however, be a great mistake to form a judgment on the nature of any colouring-matter, from the examination of its spectrum when in only one condition, or to suppose that any spectrum that may be seen is due to one single chemical compound. The effect of various re-agents must be determined, and the substances carefully examined in various ways, as described in some of Mr. Sorby's papers,* in order to distinguish or identify those which give a similar spectrum, and to ascertain whether they are mixtures or contain only one kind of colouring-matter. For this purpose a special kind of chemistry is required; and since besides this the principles involved in the study are so entirely different to those in the case of the spectrum analysis of incandescent vapours, Mr. Sorby has proposed that the name *chromatology* should be given to this subject, which should include everything requisite for the successful investigation of colouring-matters and the results derived from their study. This would form a very well-defined branch of science, sufficiently distinct from all others and wide enough to merit a special name.

Crystals.—By the aid of the spectrum microscope, we may examine extremely minute crystals, or very minute quantities of various substances dissolved in fluids. When crystals which are soluble in water are to be examined, Mr. Sorby recommends that they should be ground on moderately soft *Water-of-Ayr* stone, with a very little water. They may then be polished with a little jeweller's rouge spread upon paper over a plate of glass. Scratches on the surfaces of such crystals may be removed by rubbing them upon moist blotting-paper. Many kinds may be mounted in Canada balsam in the usual manner.

Blowpipe Beads.—The spectrum microscope may also be employed for examining substances entering into the composition of minerals, glass, &c. Mr. Sorby recommends that pieces of coloured pot mettle glass of a wedge shape, should be prepared so that the spectra of different thicknesses of known material may be compared with those of blowpipe beads coloured with an unknown substance.

Examinations of Solutions by the Spectrum Microscope.—Solutions may be examined in little wedge-shaped cells of the form represented in pl. XXIV, fig. 7. The cell may be about a quarter of an inch in depth at its deepest part, gradually diminishing to about one-fortieth. By the aid of a cell of this kind we can easily ascertain the thickness

* "Proceedings of the Royal Society," 1867, vol. xv, p. 443; "Monthly Microscopical Journal," 1871, vol. vi, p. 124. *See* also § 238.

of fluid which will give the best results. Such a cell with fluid can be placed on the stage of the spectroscope, and its spectrum compared with that produced by the object examined in the microscope.

Mr. Sorby tells me that he now preserves his solutions kept for reference in tubes of the shape and twice the size represented in pl. XXIV, fig. 6, sealed hermetically, a bubble of air being left to allow for expansion. He makes most of his experiments in cells made from barometer tubes, having an internal diameter of $\frac{1}{8}$ or $\frac{1}{6}$ of an inch, and external diameter somewhat under $\frac{1}{2}$ inch. These are cut square and ground flat at both ends, and made about $\frac{1}{2}$ inch long. They are fixed on glass with gutta percha. When filled with the liquid, the upper surface is quite level enough to enable us to examine it at once. In some cases, Mr. Sorby lays a bit of thin glass over the top, especially if he is working with alcoholic solutions or with reagents which easily oxidize. This form of cell is represented in pl. XXIV, fig. 5, half the real size. The great advantage of such cells is, that only very small quantities of material are requisite, the reagents can be added with great facility, and there is no fear of the solution running out, even when the cells are inclined. The spectra are of course modified by the presence of coloured impurities; but one great value of the spectrum method of investigation is, that many substances may be recognized with certainty, although their general colour is completely altered by being mixed with others.

Mr. Sorby recommends the pale blue solution of chloride of cobalt in a concentrated solution of chloride of calcium as the best test object for the spectrum-microscope. If two lines are seen in the orange, the definition must be very satisfactory. A very weak solution of blood is also a good test, but not for accurate definition or focal adjustment. As little as $\frac{1}{100}$ of a grain of blood in a cell $\frac{1}{10}$ of an inch in diameter, and $\frac{1}{2}$ an inch long, gives a spectrum as well marked as is possible (two absorption bands in the green)—more would cut off the whole of the green, and render the bands invisible. A solution of permanganate of potash, so dilute as to be of a pale pink colour, gives five well-marked absorption bands.

Hoppe was the first to demonstrate the peculiar absorption bands in very dilute solutions of blood, and to show that the same bands were produced in the blood of different animals. Stokes ("Proceed. Royal Soc.," 1864, vol. xiii, p. 355), proved that the colouring-matter of the blood was capable of existing in two states of oxidation, and that a different spectrum was produced according as the substance called by him *cruorine* was in its more or less oxidised condition. By protosulphate of iron, or protochloride of tin, the colouring-matter is reduced to its deoxidised state. The deoxidising solution may be made by adding to a solution of protosulphate of iron, enough tartaric acid to

prevent precipitation by alkalies. A small quantity of this solution made slightly alkaline by ammonia or carbonate of soda, is to be added to a weak solution of blood in water. By exposure to air, oxygen is reabsorbed, and the cruorine solution now exhibits the spectrum characteristic of its highly oxidised state. *See* fig. 2, pl. XXIV, A B. In venous blood part of the cruorine exists in its purple or less oxidised condition, and this in passing through the lungs becomes reoxidised and converted into scarlet cruorine.

An ammoniacal solution of cochineal gives two absorption-bands, very like those produced by blood, but they differ in relation, size and position, as may be easily proved if this and the blood spectrum be compared side by side. These two substances can be most easily distinguished by the totally different spectra obtained by adding various other reagents.

A number of other striking and well-marked spectra are described and figured in Mr. Browning's work, "Spectrum Analysis as applied to Microscopical Observation;" and also in Thudichum's Report on Chemical Identification of Diseases. London, 1868.

Preyer's work, "Die Blutkrystalle," Jena, 1874, contains a full description of some of the substances connected with the colouring-matter of blood, and gives a very useful catalogue of previous publications on that subject. In Kraus' "Chlorophyllfarbstoffe" (Stuttgart, 1872), will be found a complete history of the researches of various experimenters who have studied the colouring-matters of leaves, and a number of excellent plates which, however, in many cases represent the spectra of mixed solutions of two or more perfectly distinct substances.

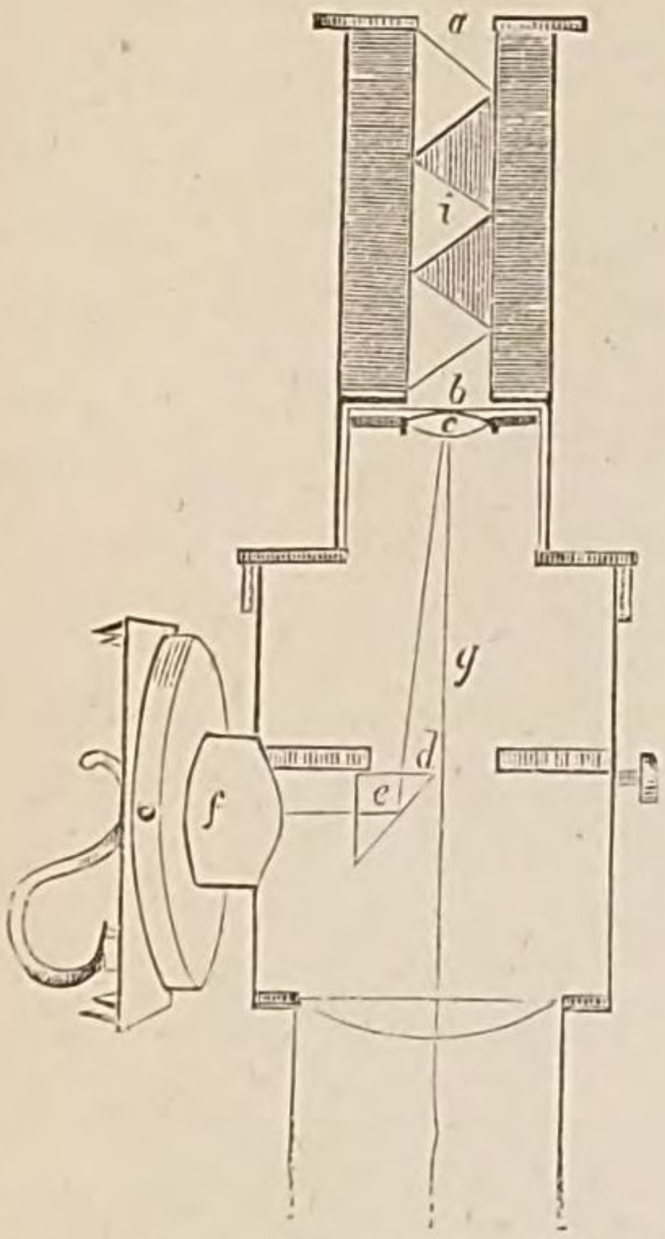
The following paragraphs have been communicated by Mr. Sorby, and contain a short general account of a number of facts which have not yet been published.

General Outline of Comparative Chromatology.

236. Application of Spectrum-Method to Coloured Constituents generally.—Since the spectrum method of inquiry can be applied to all the coloured constituents of animals and plants, it will be at once apparent that an almost boundless field is opened out for investigation. So far, Mr. Sorby has arrived at general conclusions, chiefly in the case of plants, and has described the more important facts in a paper on "Comparative Vegetable Chromatology," published in the "Proceedings of the Royal Society."* He has there shown that there are well-marked relations between the colouring-matters and the conditions under which certain plants are grown, as also between those met with in normal and abnormal varieties. The changes produced by a naturally or artificially arrested development are also very remarkable, and are, to a con-

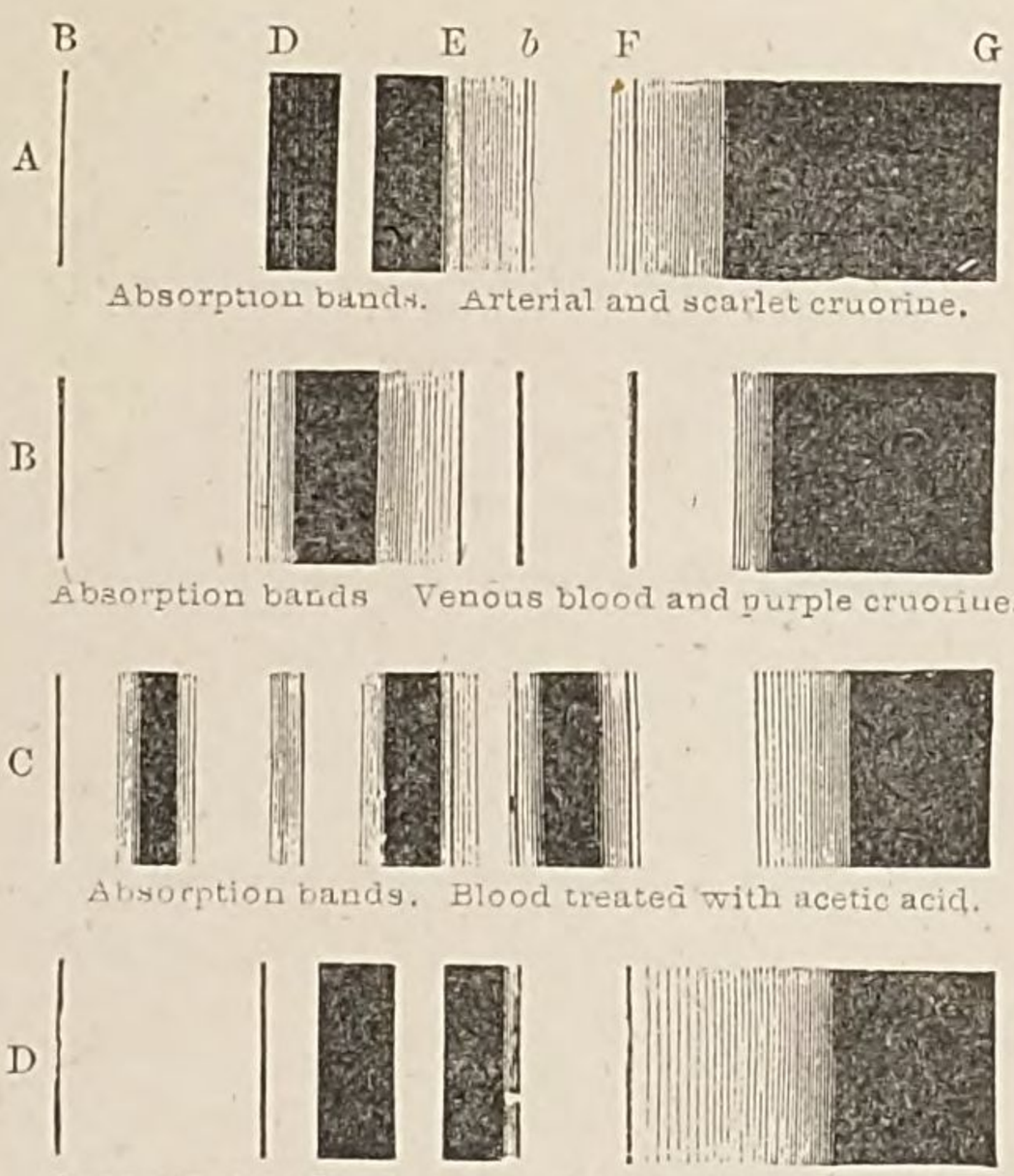
* 1873, vol. xxi, p. 442.

Fig. 1.



Mr. Sorby's spectrum eye-piece for the microscope, with arrangement for producing two spectra for comparison. p. 198.

Fig. 2.



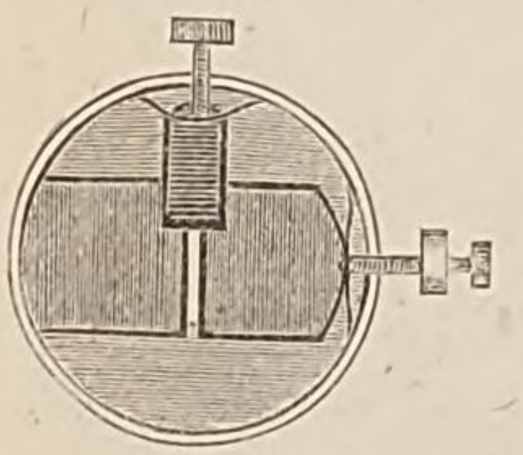
Absorption bands. Arterial and scarlet cruorine.

Absorption bands Venous blood and purple cruorine.

Absorption bands. Blood treated with acetic acid.

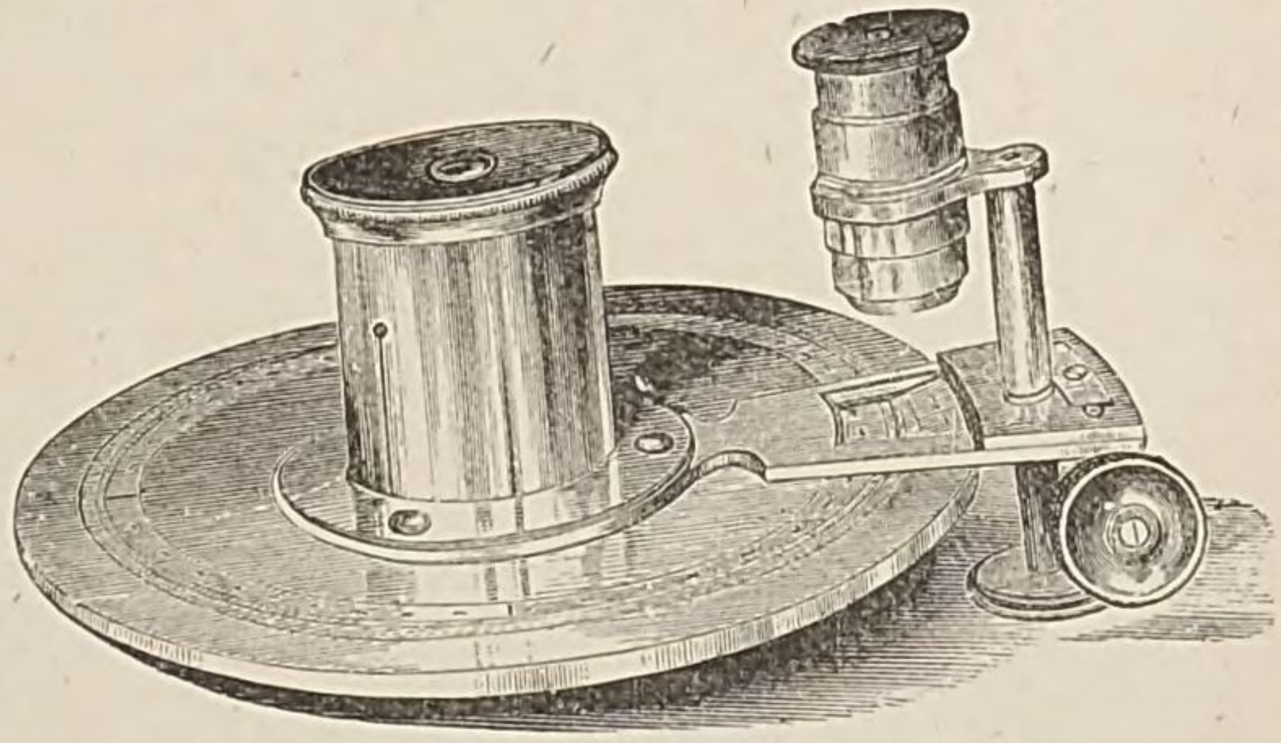
Absorption bands. Solution of hæmatin. Copied from Stokes Proc. R. S, 1864. p. 202.

Fig. 3.



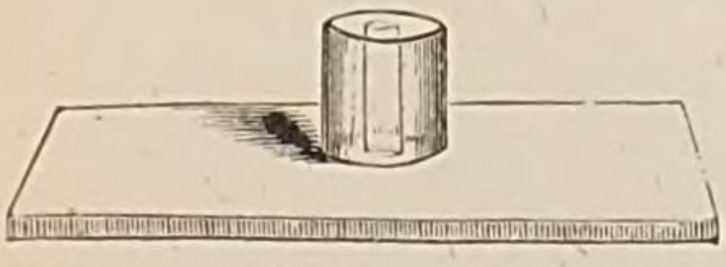
Arrangement for altering the length and breadth of the spectrum. p 199

Fig. 4.



Goniometer for measuring the angles of crystals under the microscope. p. 178

Fig 5.



Cell for examining solutions by the spectrum microscope. Half the real size p 201.

Fig. 6.

Glass tube for containing solutions for examination under the spectrum microscope. Half the real size p. 201

Fig. 7.

Wedge shaped cell, for examining solutions in the spectrum microscope p 201

[To face page 202.

siderable extent, closely connected with the optical characters of the colouring-matters, and the manner in which they are decomposed by the action of light, as is more completely described in a paper in the "Quarterly Journal of Science."* There is also a well-pronounced connexion between the nature of the colouring-matter and the position of the plants in the natural system, though the detail does not, in all respects, agree with the arrangement founded on the structure of the reproductive and other organs. As a general rule, by artificially arresting the development there is a more or less close approximation to the normal character of plants lower in the scale of general organization.

The colouring-matters found in the lower classes of animals have been studied by Mr. Ray Lankester, Mr. Sorby, and other experimenters, but, so far, no such general conclusions can be drawn as in the case of plants, because usually animals contain very few or only one kind, characteristic perhaps of a few closely related species, and not the numerous kinds met with in plants, some so characteristic of small, and others of very large groups, that their relations and differences are well pronounced. Probably, the most general results in this department of the subject are those given in Mr. Ray Lankester's paper, in which he shows the distribution of hæmoglobin through different classes of animals.† In the case of some of the lower classes of animals its place seems to be supplied by other coloured substances, which, like it, can unite with loosely combined oxygen, so as probably to be of great importance in the process of respiration. As examples, reference may be made to Lankester's "Chlorocruorin,"‡ found in the annelid *Sabella ventilabrum*, and Sorby's "Aphedeine,"§ which occurs in some species of *Aphides*. Other substances having this peculiarity, so characteristic of the chief coloured constituent of the highest classes of animals, exist in some fungi, but perhaps never in the higher classes of plants.

Mr. Sorby has also examined a number of other well-marked colouring-matters of an entirely different kind, found in the various classes of insects, and has lately studied those met with in the shells of birds' eggs, which may be called ooxanthine, oocyan, and oorhodeine. This last gives some of the most remarkable spectra hitherto discovered. The colouring of feathers has not yet been much studied, and the remarkable red substance containing copper in those of different species of plantain-eaters, described by Professor Church, is at present the only one worthy of special notice.‖

One fact that is of great importance in all these inquiries is, that the

* 1873, vol. iii (N.S.), p. 451.
† "Proceed. of R. S.," 1872, vol. xxi, p. 70.
‡ "Journal of Anatomy and Physiology," vol. iv, p. 119.
§ "Quart. Journ. of Microscop. Science," vol. xi, p. 352.
‖ "Philosophical Trans. of R.S.," 1869, vol. clix, p. 627.

exact position of the absorption bands due to any substance depends in part on the physical condition in which it occurs. If it exists in an animal or plant in a free state, the bands usually lie somewhat nearer to the red end of the spectrum than when dissolved in water, oils, or other solvents, which raise them more or less towards the blue end according to their own particular character. This circumstance enables us to ascertain the condition in which a colouring-matter occurs in living organisms, and also explains why in some cases their colour varies to some extent, even where the colouring-matter itself is in every respect the same.

237. The Spectrum-Microscope applied to Physiological Inquiries.—The application of the spectrum-microscope to physiological questions will probably lead to a number of important conclusions. Much remains to be learned, and what has been done so far merely serves to show what may be expected. Hitherto the colouring-matters of the blood and bile have attracted the most attention, but those found in urine and fæces are equally deserving of careful study; and when we come to comparative physiology, those found in eggs, hairs, feathers, scales, or other tissues open out an extremely wide field for investigation, and have even already yielded remarkable facts.

A considerable number of different coloured products may be formed by the oxidation or other changes of the hæmoglobin of blood. At least eight well-marked decomposition products can be thus formed, and it is remarkable that, like hæmoglobin itself, most of them can exist in an oxidised and deoxidised state. They can be arranged in two series, viz., one in which there are three different modifications, according as they are deoxidised, oxidised, or, so to speak, *per*oxidised, and the other in which they occur only in a deoxidised and oxidised state. The former includes hæmoglobin and products derived directly from it, and the latter hæmatin and products of its decomposition.

Since this peculiarity in uniting with loosely combined oxygen is so characteristic of products derived from hæmoglobin, and is comparatively so uncommon in the case of other animal and vegetable colouring-matters, it becomes an important character in forming an opinion as to whether one which occurs in the urine is or is not derived from the transformation of the hæmoglobin of the blood. Normal urine appears to contain two principal coloured constituents which may be called uroxanthine and urohæmin. This latter can easily be changed into a product which exists in an oxidised and deoxidised state, and very closely, if not actually, agrees with one that can be formed artificially from hæmatin. The principal colouring-matter is, however, the uroxanthine and the source from which it is derived still remains to be determined. In the case of some individuals the relative amount of these two does not vary materially, but in the case of others it varies very

considerably, and there is a very close relation between this variation and the conditions of the body at the time of the secretion. Assuming that the amount of urohæmin is constant at all times of the day, there is a great falling off in the uroxanthine during sleep and continued fasting, but a considerable increase soon after taking food, reaching a maximum four or five hours after, and then declining. There is also a rapid increase when a moderate amount of violent exercise is taken in the middle of the day, but after a certain amount there is a subsequent decrease. The effect of different kinds of food remains to be determined. The difference between maximum and minimum is far less in hot weather than in cold.

Normal healthy fæces contain a yellow colouring-matter, soluble in alcohol and in water, which, like uroxanthine, is very fluorescent, but easily distinguished from it by giving a spectrum with a well-marked absorption band. The connection between this substance and bile, and the changes which occur in various abnormal conditions, are questions well worthy of further investigation.

In jaundice, and probably in some other diseases, the yellow substance in fæces, which rapidly passes into an orange-coloured substance by exposure to the air, is not excreted in the normal manner, but occurs in large quantity in the urine in the oxidized state, and can be easily recognized by means of the well-marked absorption-band seen in the blue end of the green part of the spectrum. It is probably a product derived from bile, by some change not yet fully understood.

Mr. Sorby has studied the spectrum analysis of blood-stains with the greatest care, and, as is well known, is far in advance of any other observer in this particular branch of inquiry. I therefore gladly take advantage of his permission to print in this book the directions he has given; and as I feel that any abstract, however carefully made, might fail to convey all the necessary details of advantage to those who desire to put in practice this valuable method of inquiry, I shall only add headings to the paragraphs of Mr. Sorby's memoir, and reprint his own words without alteration. The paper itself is published in the "Monthly Microscopical Journal" for July 1st, 1871, No. XXXI, p. 9. Other methods of examination of suspected blood-stains will be more conveniently discussed when the subject of the blood is referred to. *See* Chapter XIII.

On some Improvements in the Spectrum Method of Detecting Blood. By H. C. Sorby, F.R.S., &c.

238. Spectrum Analysis of Blood.—I shall give a condensed account of what I have been able to learn in connection with this subject, and omit everything that does not bear directly on determining whether any stain is, or is not, due to *blood*. There does not appear to be any probability of our being able to decide by this means whether it is, or is not, *human*.

Spectrum-Microscope and Cells for Examining Blood.—The spectrum-microscope used in these inquiries should have a compound prism, with enough, but not too great, dispersive power, or else the bands would be as it were diluted, and made less distinct. A combination of two rectangular prisms of crown glass, with a rectangular of very dense flint, and another of less dense, of such an angle as to give direct vision, turned towards the slit, as lately made for me by Mr. Browning, appears to be the proper medium, and has other important advantages. The cells used for the experiments should be made from barometer tubing, and be about one-eighth an inch in internal diameter, and half an inch long, one end being fastened to a piece of plate glass with purified gutta-percha, like an ordinary cell for mounting objects in liquids. It is, however, a very great advantage to insert between the plate and the cell a diaphragm of platinum foil, having a circular hole about two-thirds of the internal diameter of the tube, fixed so that its centre corresponds with that of the cell. This prevents any light passing upwards that has not penetrated through the whole length of the solution, which is very important when using direct concentrated sunlight to penetrate through turbid or very opaque liquids.

Of the Reagents required.—A small spatula made of stout platinum wire, flattened at the end, is very convenient for adding small quantities of the reagents; and they should be stirred up in the cells with a platinum wire, flattened and turned up square at the end, like a small hoe. The reagents commonly employed are a somewhat diluted solution of ammonia, citric acid, the double tartrate of potash and soda, used to prevent the precipitation of oxide of iron, and the double sulphate of the protoxide of iron and ammonia, employed to deoxidize; but in some special cases diluted hydrochloric acid, carefully-purified boric acid, and sulphite of soda are required.

Varying Characters of the Stain and of the Effects of Reagents.—The character of a stain varies much with its age, and with the nature of the substance on which it occurs. If quite recent, and if the substance has no immediate influence on blood, the stain would contain little or no colouring-matter but hæmoglobin. This is easily dissolved by water, and when properly diluted—neither too strong, nor too weak—it gives

the well-known spectrum, with two dark absorption-bands in the green. The addition of a very little ammonia and a small quantity of the double tartrate produces no change, but on adding a small piece of the ferrous salt, about $\frac{1}{40}$th of an inch in diameter, and carefully stirring, so as to mix without much exposure to the air, these bands gradually fade, and are replaced by the single broad and fainter band of deoxidized hæmoglobin. When stirred up so as to expose well to the air, the two original bands of oxidized hæmoglobin can be seen again. On gradually adding a little citric acid, until the colour begins to change, these bands slowly fade away; and, if the amount of blood was considerable, a faint band would make its appearance in the red. When previously deoxidized, this solution may be turbid, but not so as to interfere with the result. The addition of excess of ammonia makes all clear again, but does not restore the original bands, or only to a slight degree, thus showing that a permanent change is produced by citric acid—the hæmoglobin is changed into hæmatin. This alone serves to distinguish blood from by far the greater number of coloured substances, which, after being changed by acid, are restored by alkalis to the original state. On deoxidizing with the ferrous salt, we obtain the well-marked spectrum of deoxidized hæmatin, with one very dark and another much fainter band in the green, almost or quite invisible when the quantity is small. If too much citric acid or double tartrate had been added, this solution might be turbid; but, if all had been properly managed, it would be quite clear. Since the deoxidization takes place rather slowly, especially in cold weather, it is well to slightly stir up the ferrous salt at the bottom, completely fill up the cell, cover it with a piece of thin glass, remove the excess of liquid with blotting paper, and mix the solution by turning the tube upside down, over and over again. On reoxidizing the solution by stirring, the bands of deoxidized hæmatin disappear, and the two bands of hæmoglobin will probably be recognized, owing to citric acid not changing the original merely into hæmatin, but also giving rise to some methæmoglobin. The whole of these facts may be seen with a single cell, containing about $\frac{1}{100}$th of a grain of blood, and any experimenter should become quite familiar with them before applying this method to suspected stains in cases of importance. Very faint bands are best seen by lamplight.

On exposure to the air in a damp place, a blood-stain may be completely decomposed by the growth of mould, but when not thus destroyed it is partly altered into hæmatin. If, however, kept dry, the hæmoglobin gradually changes into a variable mixture of methæmoglobin, hæmatin, and a brown substance not yet much studied. This change takes place far more rapidly in the acid atmosphere of towns and houses, especially when gas is burned, than in the open country; but it does occur even in the purest air, and in glass tubes hermetically sealed. The presence

of a weak acid in perspiration may also cause a stain on a worn garment to be completely changed in a very short time, and the presence of a stronger acid on dirty clothes may at once alter the hæmoglobin into hæmatin.

On digesting in water a stain that has been kept until all the hæmoglobin has disappeared, the methæmoglobin dissolves. When the solution is sufficiently strong, this shows a band in the red, and two fainter in the green. The addition of ammonia removes that in the red, makes those in the green much darker, and develops a special very narrow band in the orange. When deoxidized this solution gives deoxidized hæmoglobin. Since methæmoglobin is formed at once from hæmoglobin by the action of a great number of different oxidizing reagents, and since it can be reconverted into oxidized hæmoglobin by slight deoxidization, I am inclined to look upon it as a peculiar oxidized modification. On adding a little of the double tartrate and of the ferrous salt to even a dilute solution from an old stain, the methæmoglobin is deoxidized, and the well-marked spectrum of fresh blood can be seen. If left too long, the spectrum of deoxidized hæmoglobin is developed, but, on well stirring, that of the oxidized reappears, and the various other spectra may afterwards be obtained, as described above. That part of the stain, insoluble in water, which is chiefly hæmatin, may be dissolved in dilute citric acid or ammonia, and when deoxidized the spectrum seen to even greater advantage than when fresh blood is employed, because there is no general shading in the green, due to there having been methæmoglobin mixed with the hæmatin. We may thus obtain an excellent spectrum from a blood-stain nearly fifty years old. In very old stains all the methæmoglobin has disappeared, and sometimes even a considerable part of the hæmatin has been altered into another brown colouring-matter, which does not give any well-marked spectrum.

When a blood-stain has been made sufficiently hot to coagulate the albumen, neither water, citric acid, nor cold ammonia will dissolve it, but by heating in dilute ammonia the hæmatin is easily dissolved, and may be detected either before or after concentrating the solution by evaporation. I may here say that the spectrum of deoxidized hæmatin can in no way be better seen than by deoxidizing a solution of fresh blood that has been boiled with dilute ammonia, which gives rise to a very pure hæmatin.

Directions for Practical Examination.—In applying these principles to the detection of suspected stains, it is desirable, in the first place, to examine a portion of the unstained fabric, to ascertain whether any colour is dissolved from it by water, and whether the solution has an acid, or alkaline reaction. It is important to ascertain whether colour is dissolved from the fabric by dilute citric acid or dilute ammonia, and

if so, to determine whether this would in any way interfere with the recognition of blood by the processes described above. In the case of scarlet cloth and of some other red fabrics, much colour is dissolved out by ammonia, but not by citric acid, which ought therefore to be used, whereas in other cases ammonia is the best solvent.

Unless the stain is faint, a portion should be soaked in a few drops of water in a watch-glass, the liquid squeezed out, allowed to stand a short time in the glass, so as to deposit any small portions of the fabric, and poured into one of the experiment cells. If the stain had been recently made, and had not been changed by any special action, a solution of hæmoglobin would be obtained, and the various spectra could be seen one after the other, as already described. If, however, the stain were a few days or a few weeks old, we should obtain a mixture of hæmoglobin and methæmoglobin, or the latter alone. The various spectra could then be developed, and compared side by side with those from fresh blood, to be sure that there is complete correspondence in the position and relative intensity of the bands. The residue insoluble in water should then be dissolved in dilute citric acid or ammonia, according to the nature of the fabric, and the spectrum of deoxidized hæmatin developed. If insoluble in cold citric acid or ammonia, hot ammonia should be tried, since the stain might have been so heated as to coagulate the albumen. If it be desirable to keep the specimen of deoxidized hæmatin for subsequent reference, the cell may be covered with a piece of thin glass, and after removing the excess of liquid, the edge of the cover painted round with gold-size. When properly managed, such an object will show a perfectly good spectrum, even after many weeks.

Of the most important Spectra peculiar to Blood.—If therefore we have a sufficient amount of a moderately old stain, we may easily see in succession the seven very different spectra of the following solutions :—1. Neutral methæmoglobin. 2. Alkaline methæmoglobin. 3. Deoxidized hæmoglobin. 4. Oxidized hæmoglobin. 5. Acid hæmatin. 6. Alkaline hæmatin. 7. Deoxidized hæmatin. If the amount was very small, only Nos. 4 and 7 would show distant bands, and the rest would be characterized rather by their comparative absence ; and it must always be borne in mind that Nos. 1 and 2 may be modified by the presence of unaltered hæmoglobin, No. 3 by that of dissolved hæmatin, and Nos. 5, 6, and 7 by that of undecomposed hæmoglobin or methæmoglobin.

It would be easy to obtain other preparations, and to see several other spectra derived from blood, but it appears to me unnecessary, since the above are so remarkable and unique in the manner in which they are produced, one after the other, especially deoxidation and re-oxidation on stirring, which seldom occurs in other colouring matters,

that they afford as satisfactory a test for blood as could be desired, and still more so when we consider not only the general character of the spectra, but also the exact position of the absorption-bands, and that some are unusually distinct.

Precautions necessary in Examining very faint Blood Stains, &c.—The above directions apply to simple cases, where the amount of material at command is amply sufficient, and the fabric on which the stain is found does not contain anything that makes the blood insoluble, or interferes with the various tests. I shall, however, now describe what should be done in cases which are made specially difficult by various causes. If the stain were very faint, from the presence of very little blood, or if the greater part had been removed by washing with water, it might be desirable not to divide the material, but to examine the whole at once. The stained portion should therefore be digested in a few drops of dilute citric acid or ammonia, and the presence of hæmatin determined, as already described. If faint and spread over a considerable surface, it might be well to digest in citric acid or ammonia diluted with much more water than would fill the experiment cell, and afterwards concentrate the solution by gentle evaporation. By this means blood could be detected, even when considerable effort has been made to remove it, and only a faint brown tinge left, just visible on white linen. There would generally be no difficulty in the case of a stain on cloth which had been sponged, for enough blood solution would be left in the fabric.

The effects of Mordants on Blood Stains.—The presence of mordants in cloth or prints may require us to somewhat modify our proceedings, especially if the stain had been made wet, and to a great extent removed, so that we have only the dried-up solution of blood, thoroughly incorporated with the mordant. Certain kinds of brown cloth are of such a character, and about seven years ago portions of a wetted stain were sent by me to a number of the highest authorities in the detection of blood, and they said that neither they nor anyone else could recognize it. However, by proper care, I found that after a lapse of six years it could be detected by the spectrum method. The best plan was to digest a portion of the cloth in dilute ammonia, and to squeeze it well over and over again, with a pair of forceps, and finally with the finger and thumb, so as to obtain as much of the solution as possible. This was very turbid, but when deoxidized in the usual manner, and illuminated by concentrated light direct from the sun itself, the band of deoxidized hæmatin was quite distinct. When the cell was kept for a while, so that the insoluble part settled to the side, no band was visible, and therefore the hæmatin was evidently combined with the mordant. It will thus be seen that it may be most important not to filter or allow the insoluble matter to subside, but to overcome the opacity by means of a sufficiently

intense light. If the sun could not be made use of, the lime or electric light would no doubt be the best substitute.

Effects of Vegetable Soil on Blood Colouring Matters.—When fresh blood solution is agitated in a test-tube with vegetable soil, and left until quite clear, the colouring matter is completely carried down with the earth. Dilute ammonia, however, dissolves out hæmatin, and therefore, in testing portions of soil, they should be digested in considerably more of that solvent than will fill an experiment cell, and after the solution has become quite clear it should be concentrated by evaporation. The spectrum of deoxidized hæmatin may then be seen by following the ordinary method. The same process should be adopted in examining stains on clothes impregnated with earth or earthy dust, and marks on iron contaminated with much rust, if water will not dissolve out unaltered blood or methæmoglobin.

Of Detecting Blood Stains on Leather.—The importance of being able to detect blood stains on leather was prominently brought before me by a case in which the trial of a suspected person depended on the nature of certain dark marks on his gaiters. The presence of tannic acid so completely mordants the blood, that neither water nor citric acid will dissolve it, and ammonia gives rise to a most inconveniently dark solution. If the stain is on the surface, and has never been wetted, a thin shaving should be cut off, so as to have as much blood and as little leather as possible, and the blood should be dissolved off without exposing the solution to the action of the leather itself. This may be accomplished by taking one of the experiment cells, nearly filled with water, bending the shaving, and inserting it in the upper part of the tube, so as to touch the water, being careful to arrange it so that the stain may be on the convex side of the leather, and in contact with the water. When a drop of blood falls on leather, many red globules are filtered out from the serum and left on the surface, and, when thus treated, they dissolve, and the coloured solution sinks at once to the bottom of the cell, without coming in contact with the leather. The various spectra may then be observed in the usual manner. This method would be of little or no use if the stain had been wetted, and for a long time I concluded that after such treatment it would be impossible to recognize blood. However, after many experiments, and after having again and again almost given up the inquiry in despair, I found that the difficulty could be overcome in a very simple manner. The best solvent for the insoluble compound of the colouring matter of the blood with tannic acid, is hydrochloric acid diluted with about fifty times its bulk of water. If stronger or weaker, the result is not so good. When a portion of unstained common brown leather is digested in this dilute acid, the solution is scarcely tinged yellow. On adding excess of ammonia, the colour becomes pale purple, or neutral tint, made deeper when the

double tartrate and the ferrous salt are added, but remaining nearly clear. This gives a spectrum very dull all over, but without any trace of definite bands in any part. The depth of colour varies much with different specimens of leather. A portion of similar material soaked with wetted blood, gives a yellow solution, made brown-purple and turbid by the double tartrate and ammonia, and remains so when deoxidized. The band of deoxidized hæmatin can however be distinctly seen with a light sufficiently strong to penetrate the turbid and dark solution. Before examining the suspected stain, it would be well to make out how much of the unstained leather could be used without giving too dark a solution, and to use no more of the stained. If the deoxidized solution be too turbid, the cell may be kept for a while horizontal, until the deposit has subsided sufficiently to allow the principal absorption-band to be seen; but it is not so distinct, when all have subsided, as though the greater part of the hæmatin still existed as a compound insoluble in dilute ammonia.

The presence of tannic acid in wood and other substances might make it necessary to employ a similar process, if the relative amount of blood were so small, that none could be dissolved out by water, or dilute citric acid.

Of preventing the Injurious Effects of other Colouring Matters.—Cases might occur when it would be necessary to decide whether blood were present, along with some other coloured substance, soluble in water. The method to be employed would depend much on the nature of this impurity. If it were a colouring matter, belonging to what I have described in former papers as group A, in which the absorption is removed by sulphite of soda, in an alkaline solution, there would be no difficulty in seeing all the spectra. Thus, for example, it is easy to add so much magenta to the solution of a little blood, that its absorption-bands are entirely hid; but a small quantity of sulphite of soda so completely removes the colour of the magenta, that the various spectra of the blood may be seen almost as well as if it had been pure.

The colouring matters of my group B that are most likely to occur, are those of fruits, and in them the presence of the free acid would be almost certain to have changed the hæmoglobin into hæmatin. The best plan would then be to add excess of ammonia, and, if the solution were made too dark, to dilute it with so much water that the strongest light at our command would show the green part of the spectrum sufficiently bright to prove that no absorption-band occurred there. On deoxidizing in the usual manner, the solution may be made somewhat darker by the presence of tannic acid, but the darker band of deoxidized hæmatin could be recognized without material difficulty.

By far the greater number of the colouring matters belonging to my group C are yellow and orange-coloured; and since these chiefly absorb

the blue rays, they do not interfere with our seeing the bands of the blood spectra, which occur in the green. Cochineal is one that requires special attention. The addition of ammonia to its solution in water gives rise to two bands in the green, which, though differing materially from those of blood, are yet so nearly in the same situation, that they completely disguise the presence of a small amount of blood. However, on adding a small excess of boric acid, the bands of the cochineal are made more faint, and very considerably raised towards the blue end, so as to leave the red end of the green clear, whilst those of oxidized hæmoglobin are not changed, and that nearer the red end, if not both, can be seen perfectly well. By proceeding in the usual manner, there is no great difficulty in recognising the darker band of deoxidized hæmatin.

Undecomposed Hœmatin, if present, may always be Detected.—Other special difficulties might occur in particular instances, but I trust that these examples will suffice to show how they may be overcome. I do not now know of any that require special remarks; and, as far as I am able to judge, we need never despair of detecting blood, so long as any hæmatin remains undecomposed. Fortunately it resists decomposition so well, that this would rarely happen in ordinary circumstances; but yet there are cases in which it does occur, as, for example, when acted upon by strong ozone, or other powerful oxidizing reagents.

It is quite possible that stained garments might have been washed, and some of the water employed might be obtained for examination. If no soap had been used, this water could be examined in a long tube of thick glass, ten inches or more in length, and a quarter of an inch in internal diameter, permanently closed at one end with a circular piece of plate glass, and, when filled, covered over at the other with another glass. For examining solutions in such tubes a small pocket spectroscope, such as recently made for me by Mr. Browning, is extremely convenient, and suitable in every respect. If only two or three days old, the bands of oxidized hæmoglobin might be seen; but if the solution had been kept longer, and they could not be detected, it should be concentrated by evaporation at a gentle heat, and tested for hæmatin. If during evaporation any deposit be formed, insoluble in cold dilute ammonia, it should be dissolved by the aid of heat. When soap is used in washing off stains, the alkali soon changes the hæmoglobin into hæmatin, and the soap makes the solution inconveniently turbid and opaque. It is best in such a case to agitate the suspected soap and water with ether, remove it with a pipette, after the two liquids have completely separated, and repeat the process over and over again, with fresh ether, until the aqueous solution at the bottom has become quite clear and free from soap. It should then be concentrated by evaporation, and examined for hæmatin, as usual. Of course in all such cases

it would be desirable to test the solution as soon as possible, lest decomposition should occur, but by these means a very small quantity of blood, that would show no colour, might be recognised within a week or two, but probably not after.

Method of Detecting Blood in Urine.—For the detection of blood in urine, a tube about ten inches long is very suitable. If turbid it should be filtered; but, since a considerable number of red globules might be separated, the deposit on the filter should be dashed with a little water, and this solution either examined by itself, or added to the filtered urine. If the depth of colour in the ten-inch tube be so great, that the yellow end of the green part of the spectrum is absorbed, the urine must be somewhat diluted, or examined in a shorter tube. When the depth of colour is about an average, I find that by this means as little as $\frac{1}{10000}$th part of blood can easily be detected in fresh urine, which is equivalent to about one drop in a pint.

PART II.

THE MICROSCOPICAL CHARACTERS OF THE SIMPLEST PARTICLES OF TISSUES AND OF THEIR DEMONSTRATION—OF STRUCTURAL ELEMENTS AND ELEMENTARY PARTS IN HEALTH AND DISEASE—OF DEPOSITS FROM FLUIDS—OF ANIMAL AND VEGETABLE PARASITES.

CHAPTER XII.

Of the Simplest Particles and Anatomical Elements met with in the Body in Health and Disease. — Granules. — Fatty Granules. — Albuminous Granules. — Pigmentary Granules. — Earthy Granules. — Globules. — Air-bubbles. — Oil-globules. — Albuminous Globules. — Earthy and Crystalline Globules. — Corpora Amylacea. — Of Fibres. — Of the Bioplasm or Living Matter of the Body and of Elementary Parts or Cells. — Of the Structure of Cells. — Of the different kinds of Cells and Elementary Parts. — Of Epithelium: scaly, tessellated, glandular, columnar, ciliated. — Of False Cells. — Demonstration of Cell Structures. — Demonstration of Bioplasm. — Of Fibres. — Of a Fibrous Appearance. — Capillaries. — Of the Changes of the Elementary Parts in Disease.

In this chapter I propose to direct attention to particles of various kinds which may be found in the tissues and fluids of the body in health and disease. The particles in question varying much in size, form, composition, and proportion, have not been very accurately described. Names given to them have not always been chosen with due care, and the same name has even been applied to bodies essentially different from one another, while things of the same nature, but discovered in different situations, have been spoken of under more than one name. This question of nomenclature is of great importance, and every one using the microscope should make himself familiar with the terms employed by the most experienced practical observers when describing the facts previously observed by them.

If doubt exist in the observer's mind as to the precise meaning of a term he feels constrained to employ, he should give in a note the definition which he considers to be correct, or he should describe in detail the appearances which have been observed by him.

As an attempt has been made to explain the meaning of some of

the terms frequently used by microscopists and often recurring in this work, it is hoped that the reader will not too hastily glance over the sections below.

Of the minute particles to be considered some are of structural or functional importance, some are accidental in their presence, while some indicate that certain changes have taken place in tissues and important organs which render impossible further healthy action. Amorphous granules, minute crystalline particles, globules differing much in form, size, and refractive power may be present in a texture, which is quite destitute of them in health, or may be suspended in fluids which ought to be perfectly free from such particles. Again, actual fibres distinct from one another or collected in bundles, or a "fibrous appearance" due rather to the manner in which a tissue has been formed than to actual fibres present, may be observed in young tissues which ought to be clear and transparent and without any indication of structure in a healthy condition. Such appearances, in some cases, indicate that a texture has grown old before its time and this practically may be looked upon as a morbid change.

Tissues remarkable for their glass-like transparency in health, often become in the course of disease more or less opaque, and the change may be due to the deposition of *granules*, *globules*, or *fibres*, or to the conversion of normal structure into insoluble or sparingly soluble granular matter.

But besides the presence of non-living particles which may have been deposited from nutrient fluids or formed in the course of unusual chemical changes in the elements of normal structure, tissues may be rendered less transparent than in the normal state, and fluids may be made almost opaque by the presence of multitudes of *minute living bioplasts* which have very quickly grown and multiplied. Indeed this, I am sure is the most common though the least recognised cause of tissue degeneration. It generally constitutes the first change in the morbid process, and the non-living *granules*, *globules*, and *fibres* afterwards seen result from the death of the *minute living bioplasts*, and the deposition of compounds produced at the moment of death, or afterwards. The nature and origin of the living particles of bioplasm is very different in different cases. Sometimes they are bacteria germs or living particles (microzymes) which constitute the first stage of development of certain microscopic fungi, but more often they are minute bioplasts which have originated in the already existing bioplasm of some part of the organism which placed under unusual conditions as regards the supply of nutriment, has grown and produced a form of rapidly growing bioplasm which may merit the term *abnormal* being applied to it.

From one or other of the above-mentioned causes, or from a combina-

tion of several, changes may be so great as to cause the natural structure of the tissue to be completely obscured, though it may not have been completely destroyed. If the thinnest possible section be submitted to examination, no ordinary structure may be discerned, the whole appearing confused and indistinct.

In like manner the normal elements suspended in a fluid may be obscured by adventitious particles which are not usually present. I have seen capillaries packed so full of minute particles that not a single blood-corpuscle, nor the nuclei (bioplasts) of the capillary wall could be seen. Now if we desire to ascertain the actual nature of the particles in tissues and fluids, in many cases we must endeavour to make more transparent the granular material which gives the whole its confused appearance and opacity, in order that we may study the arrangement of the new matter and ascertain whether there be any actual remains or indication of the normal structure left. This clearing may be effected in two ways :—1. By the action of a fluid which refracts more highly than water or serum. 2. By the addition of some chemical reagent which is known to have the property of dissolving or much altering the substance which gives rise to the opacity.

The use of highly refracting fluids in microscopical investigation has been already referred to in pages 57, 58, and the influence of chemical reagents, and the methods of applying them have been discussed in the early part of Chapter X, page 155.

239. Of the simplest Particles and Anatomical Elements met with in Health and Disease.—No accurate description of anatomical changes, no clear account of the phenomena of living healthy and morbid textures can be given until the meaning of the term used has been clearly defined. Want of attention to this point has not only occasioned great perplexity but has led to such confused ideas that we can scarcely hope that anything like a clear account of some of the simplest morbid changes will be given in our text books for many years to come. The student must think for himself and must think independently. In many departments not only will he be misled by authoritative statements, but he will find authority, misleading and being misled by authority, as if authorities had agreed not to be too hard upon one another in order that a measure of credit might be enjoyed by each. And until it is generally considered that authorities ought to define what they mean by the terms they use, confusion of ideas will prevail with reference to many things that might be made clear enough.

If an observer is in doubt whether the character of the object is truly conveyed by the words and phrases he selects, a note of interrogation should be placed in brackets to indicate doubt, or the sense in which the word is employed should be explained in a note. Whenever our meaning can be rendered clear to others by drawings, these should

invariably be appended. *See* remarks upon the importance of drawing in page 33.

240. Granules are minute bodies of no determinate shape or size. They may seem to be only separate dots or points when examined by the highest powers of the microscope. They cannot be measured. When granules are deposited in a tissue, the change may be described as "a granular appearance." When suspended in limpid fluids, granules or minute particles of matter manifest peculiar movements which are dependent either upon currents in the fluid caused by the gradual evaporation taking place from the surface, or from the edges in cases where the drop is covered by thin glass,—upon the operation of the force of gravitation acting upon the particles themselves,—upon communicated vibrations,—or upon electrical disturbance. The movements referred to affect alike particles of organic and inorganic matter—lifeless, as well as living particles. The movement was first carefully studied and described by Robert Brown, the botanist, and was termed by him *molecular motion.* The particles are often called *molecules.* Molecular movements may be seen in the chyle, in urinary deposits consisting of urate of soda in a state of minute division, and indeed wherever fine particles of matter are suspended in a fluid. These movements are frequently observed in the interior of cells where insoluble particles are suspended in a limpid fluid. In the interior of many "pigment cells" as those in the frog's skin and in the choroid coat of the eye molecular movements may be observed.

Granules may be divided according to their composition into three principal classes, *fatty granules*, *albuminous granules*, and *earthy granules.* It is impossible to distinguish these from one another by their microscopical characters alone, and it is therefore necessary to resort to chemical analysis. For this purpose, the granular matter suspended in water is placed under thin glass in the usual way, but in order to obtain a sufficient thickness of fluid for examination, it is desirable to prevent the thin glass from coming in too close contact with the glass slide, by inserting a piece of hair or hog's bristle. The slide being placed under the microscope, a little of the reagent is forced out of one of the tubes with capillary orifices, § 138, upon the slide, so that it may gradually pass in between the glasses, while the effect it exerts upon the granules may be studied under the microscope.

Particles, which would be correctly termed "granules" when subjected to examination by low powers, may exhibit definite forms under the highest magnifying powers, in which case the term "granule" is not applicable. The granules in pl. XXV, fig. 13, when highly magnified, appear as shown in fig. 6, and assume the form of "globules." Others again still retain their indefinite granular character under the highest magnifying powers as those in fig. 3, but they would still be termed

"granules." Some "granules" become resolved into well-defined crystals when submitted to examination with the aid of the $\frac{1}{25}$ or $\frac{1}{50}$.

Granules consisting principally of Living Matter.—Although living matter often appears "granular," the matter which is actually alive is perfectly clear, transparent, and structureless. It has been said, that living matter is only to be distinguished from non-living albuminous matter by its "granular appearance," but the statement is altogether erroneous, and results from most careless observation, if indeed it results from observation at all. It so happens that a "granular appearance" is not a characteristic of *living matter*. Many kinds of matter not living are "granular." Comparatively few specimens of granular matter are living. Most of the "granules" in living matter are lifeless, and of the so-called living "granule" the greater portion is not alive. The outer part of the bacterium which gives to that body its characteristic appearance is not living, and the minute particles constituting bacteria germs and the so-called microzymes and micrococci, collections of which form what has been termed granular matter, and which have been erroneously supposed to form higher forms by coalescence, are not wholly alive. It is now nearly fifteen years since I first called attention to the characters of living matter or bioplasm, and to the appearances observed when minute particles of living matter were placed under powers magnifying upwards of seventeen hundred diameters.

Fatty Granules.—Fatty matter in a granular state is found in health in the chyle, and sometimes in the blood, and in many tissues and fluids in disease. These granules are very often minute—so small, indeed, that they pass through the interstices of the most delicate membrane and also through those in various kinds of formed material. They are not affected by *acetic acid*, but are often dissolved or saponified by an *alkali*. They are readily dissolved by ether, and as the ethereal solution evaporates, fat in the form of globules, often of considerable size, remains behind. Fat "granules" often appear as minute "globules" when highly magnified.

It is probable that many granules consist of a combination of fatty and albuminous matter. Much of the fatty matter in a granular state, which is suspended in albuminous liquids, deposited in tissues, forming the contents of cells, or resulting from the disintegration of tissues, contains a large quantity of cholesterine, which is easily extracted by treating the substance with alcohol, § 226. By evaporating the alcoholic solution, the cholesterine will be obtained in a crystalline form.

Albuminous Granules.—By this term I wish to imply all lifeless granules composed of any modification of albumen, fibrin, casein, or other substance belonging to this class. These granules are found in many of the cells of the healthy organism, and in a vast number of

tissues at all periods of life. In the early stages of development, "granules" are very abundant, but many of these consist of particles of living bioplasm; but interspersed are particles which have resulted from the death of bioplasm. Albuminous granules are often suspended in fluid. They are usually soluble in acetic acid—always so in the earlier period of their formation. They are also soluble in alkalies. Ether has no effect upon them.

Pigmentary Granules are found in abundance in the cells of the choroid coat of the eye, pl. XXV, fig. 8, in the cells constituting the deeper layer of the epidermis, in the hair bulbs, in the bronchial tubes, in the cells composing melanoid cancer and various morbid growths, and in other situations. Their character may be studied in the pigment cells of the skin and coats of the blood-vessels of many batrachia, as the frog (pl. XIII, fig. 4, p. 144), and newt, and many fishes. These pigmentary granules are formed from the bioplasm matter of the cell. They may be removed from the cell, and when they escape into the surrounding fluid they exhibit molecular movements. Under the highest powers they exhibit no definite form. The dark granules often found in sputum forming irregular masses, embedded in mucus, and appearing as if inclosed in a membrane, seem to consist in many cases merely of blacks which have been inspired, but in others probably of pigmentary matter formed in the lung itself. Urates of soda and ammonia are often precipitated as granules which are soluble in hot water, pl. XXV, fig. 1.

Earthy Granules are also widely diffused in the animal body, deposited in solid tissues and suspended in the fluids. In old age, many tissues are largely impregnated with granules consisting of earthy matter. They may consist of phosphate or carbonate of lime, phosphate of ammonia and magnesia, and more rarely of other earthy salts.

If composed of carbonate, they effervesce upon the addition of an acid, and readily dissolve. If they dissolve without effervescence, and the clear acid solution yields with ammonia a precipitate in a *granular state*, phosphate of lime is present; if *crystallized*, triple phosphate.*

241. Globules.—A "globule" is more or less spherical in form. Globules vary much in size, and, like granules, differ in their chemical composition, as well as in other characters. Some are composed of albuminous matter, others consist of fat. Phosphate and carbonate of

* For the method of applying the test, see page 168. There is a possibility of error when a fluid or tissue in which the granules are deposited, contains carbonate of ammonia from decomposition. This salt, however, can always be very readily removed by the addition of water in which it is readily soluble, in the first instance. If the deposit which effervesces has been heated to redness, it cannot of course contain carbonate of ammonia. It must, however, be borne in mind that when salts of many of the organic acids, as citric, oxalic, lactic, acetic, &c., are incinerated, *carbonates* are found in the ash.

lime, and other mineral matters are the materials of which many globules are composed. The appearance of the globule, when examined by transmitted light, varies according to the medium in which it is placed. If the globule and the medium are colourless, and exactly, or nearly correspond in refractive property, the globules may be invisible, but if the globules and the medium differ much in refraction, the outline of each globule will appear dark and well-defined, and its centre clear and bright. The width of this dark outline is determined by the difference in refracting power. For instance, the outline of an oil globule in water is distinct and well defined, but narrow, pl. XXV, figs. 6, 9, 10, 11, while the outline of an air bubble in water is very much wider than that of the oil globule, fig. 5.

If the globule is suspected to consist of an earthy material, it must be tested with chemical reagents. Phosphate of lime is readily dissolved by acids, without effervescence, and may thus be very easily distinguished from fatty matter, while the latter is dissolved by ether, which has no action whatever on the former.

Considerable confusion has been occasioned with reference to the terms *granule*, *molecule*, and *globule*. By some writers the two former have been used in the same sense as "globule." The latter word should, however, be restricted to a body which has a distinct circular outline, with a clear bright centre; while by "granule" is understood a minute particle of no determinate form. The latter is, therefore, synonymous with the word "molecule." It seems to me very important that we should carefully distinguish the mere *molecule* or *granule* from the well-defined *globule*. We can discover the form of a globule without difficulty, but are quite unable to ascertain that of a granule or molecule.

Globules consisting partly of Bioplasm.—Among the best-defined "globules" are the spores of fungi with their dark well-defined circular outline and clear transparent centre. A sporule of the yeast fungus or penicillium glaucum is a good example, pl. XXV, fig. 7. The dark outline is caused by the highly refracting capsule composed of formed material. This contains the perfectly clear structureless living matter or bioplasm to which the phenomena of growth are entirely due. This important living matter has been entirely passed over by many observers, and by some has been considered to be mere passive fluid, holding certain materials in solution, but not endowed with vital properties. So far from being considered to be actively concerned in growth, it has been looked upon by many as *passive*, and, as compared with the capsule, *unimportant*.

Air-bubbles.—The student should familiarise himself with the character of air-bubbles as they appear in various fluid media of different refracting power, or he will assuredly make the most ridiculous and unpardonable mistakes. Air-bubbles and oil-globules should be examined

in spirit, water, glycerine, Canada balsam, and other media, as well as under various powers of the microscope. Air-bubbles can always be obtained very minute, by placing a drop of gum water on the glass slide, and raising and depressing very rapidly a piece of thin glass which is well wetted with it.

Oil-globules may be easily obtained for examination by shaking a few drops of oil in a bottle of weak gum water. Or a drop of milk in which they exist ready formed of all sizes, may be placed under the microscope, pl. XXV, fig. 10.

Globules composed of Fatty Matter are so frequently met with in the healthy organism and in morbid conditions, that every one must have observed them. They are found in health in the liver cell, and in the epithelium of the small intestines in considerable number, in the cortical portion of the suprarenal capsules, in the cells of sebaceous follicles, and in those of the mammary gland, in the marginal tufts of the placenta towards the end of the period of gestation, in the muscular fibre-cells of the uterus after delivery, and in the cells which are found in considerable number in the colostrum, or first portions of milk secreted by the mammary gland each time it is called upon to discharge its function. The so-called "nucleolus," it has been said, usually consists of an oil-globule, but a true nucleolus is a new centre, and consists of living bioplasm, not of non-living oil. *See* p. 226. In morbid conditions there is not a tissue in the body which may not become studded with oil-globules. Even the transparent cornea, vitreous humour, and crystalline lens are not free from them, nor is there a fluid in which they do not sometimes occur. In disease, fat-globules are often found in epithelial cells, especially those of the liver, kidney, and many other glands, in muscular tissue, in nerve, fibrous tissue, cartilage, basement membrane, as of the lung in emphysema, in the cells of mucous membranes, and in those of the bronchial tubes in catarrh, in inflammatory exudations generally, in the fluid which collects in serous cysts, and in certain cavities as the antrum, and in many other situations which will be enumerated in their proper place. When free to move in fluid, minute oil-globules become aggregated together to form collections or masses which have often been mistaken for *cells*, pl. XXV, fig. 11. This aggregation is a physical phenomenon, and depends upon the attraction of gravitation.

The deposition of oil-globules seems to be constant wherever a tissue ceases to discharge its office, either in the natural course, or in consequence of a morbid process having been set up. The deposition of fat-globules, or more probably the *conversion* of albuminous material into fatty matter, appears to be a natural change prior to the absorption of many tissues. Albumen and fibrine if exposed to the prolonged action of air and water will yield among other products fatty matter which takes the form of globules, and in the disintegration of

PLATE XXV.

Fig. 1.

Granules of urate of soda. p. 220.

Fig. 2.

Granules under a power of 215. p. 218.

Fig. 3.

Granules highly magnified. p. 218.

Fig. 4.

Granules, consisting of fatty matter in a state of very minute division, from 'chylous' urine, resembling the 'molecular base of the chyle.' Some small cells are also observed. × 215. p. 218.

Fig. 5.

Air bubbles in water. p. 221.

Fig. 6.

Part of fig. 2 more highly magnified. The granules become globules. p. 218.

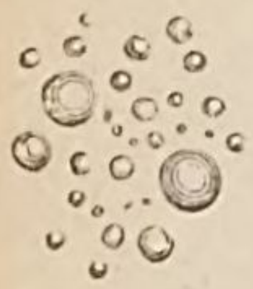
Fig. 7.

Sporules and Mycelium of sugar fungus, after Hassall. p. 221.

Fig. 8.

Pigment cells from the outer part of the choroid. Human. × 150. p. 220.

Fig. 9.

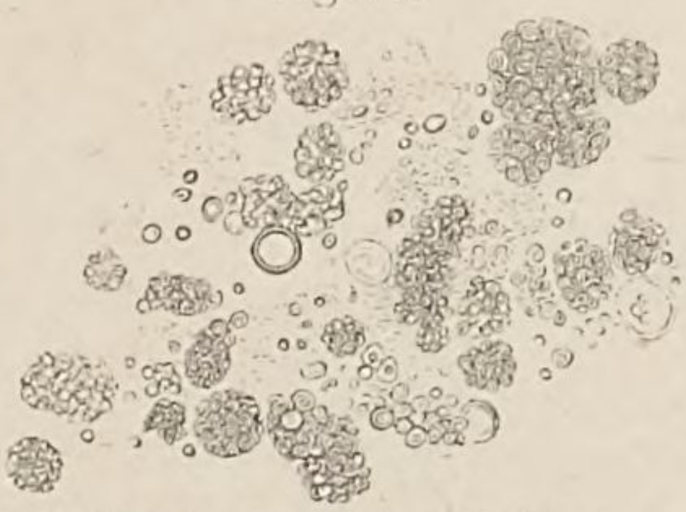


Free oil-globules. p. 221.

Fig. 10.

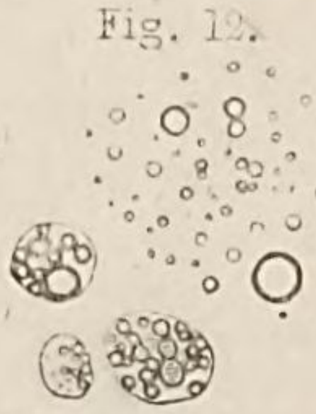
Oil-globules from Milk. × 215. p. 221

Fig. 11.



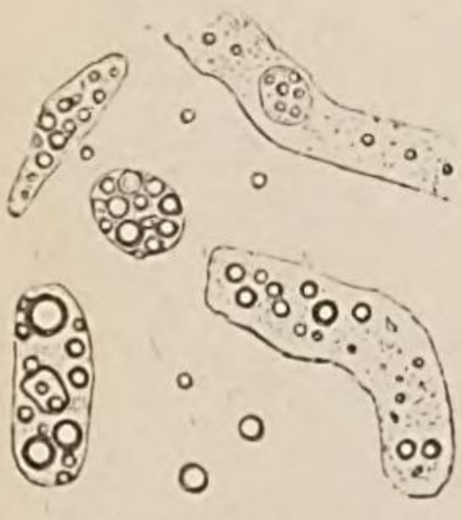
Collections of minute oil-globules, the so-called inflammation corpuscles, compound granular corpuscles, &c., from softened brain. p. 222.

Fig. 12.



Oil-globules, free and enclosed in cells. × 215. p. 218.

Fig. 13.

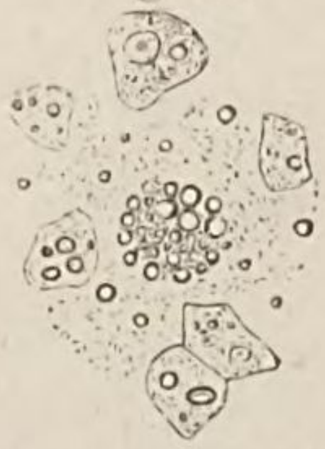


Casts of the uriniferous tubes, containing free oil-globules and cells filled with fatty matter, from a case of fatty degeneration of the kidney. p. 218.

Fig. 14.

Oil-globules *a*, and granules *b*, enclosed in cells.

Fig. 15.



Oil-globules in the so-called cells of the liver; in the centre is seen a collection of oil-globules not surrounded with any envelope. × 215. p. 218.

Fig. 16.

Organisms met with in urine. *a*, different forms of fungi. *b*, Bacteria, or vibriones. × 215. p. 219.

Fig. 17.

Penicillium glaucum, in different stages of development, from acid urine. × 103. p. 221.

Fig. 18.

Corpora amylacea, from the brain. p. 224.

1000th of an inch ⌊___⌋ × 215.

" " ⌊_____⌋ × 400.

[To face page 222

tissues in the living body fatty matter is formed, but in health this oily matter is absorbed, as fast as it is produced. In some conditions its removal is interfered with or rendered impossible. If the due activity of a tissue is impaired by disease, the development and deposition of fat-globules often ensues. If the morbid alteration be limited in extent, while the system generally remains vigorous, the absorption of the oil-globules and other products of the degenerated tissue may slowly follow; but if the patient's strength be reduced from any cause, the degenerative process continues, the functions of organs necessary to life soon become deranged, and death follows before long. It is in such conditions of system that maladies and trifling surgical operations, from which healthy men would recover, prove fatal.

It is important to bear in mind that some forms of fungi closely resemble oil-globules in their general appearance. They do not, however, refract the light so highly as the latter, while acetic acid generally renders them more transparent. On the other hand, ether has no action upon fungi, but it dissolves the oil. The outline of small oil-globules is relatively thicker than that of large ones; but the thickness of outline of fungi does not vary in this manner. Fungi are represented in pl. XXV, figs. 7, 16, 17. Generally, oil-globules in a specimen vary very much in size, while the round sporules of fungi if not all of the same dimensions, approach nearly to one standard. Practice and experiment, however, will enable the student to accurately distinguish oil-globules from fungi.

Non-living Globules composed of Albuminous Material are often of a very large size; they are found in many morbid products, such as serous fluids from cysts (pl. XXVI, fig. 1), soft malignant growths, and they have been detected in the eye in disease of the choroid and retina. The outline of such globules is an exceeding fine line, and the globules themselves are scarcely visible, except under the influence of a very dull light. Some of the bodies above referred to have been termed *colloid* bodies.

Globules composed of Earthy or Crystalline Matter.—Hard globules consisting of earthy matter, may have a composition similar to that of certain granules composed of inorganic substances. Many large globules met with in the brain substance and in the choroid plexuses, pl. XXVI, fig. 26, contain a large quantity of carbonate of lime, while those which occur in the urine of the horse and of rodent animals, figs. 3 to 6, are composed entirely of this substance.

Many calculi at an early stage of their formation might correctly be termed *globules*. By the gradual deposition of new material externally, globules originally microscopic, may attain a very large size. *See* "Kidney Diseases, Urinary Deposits, and Calculous Disorders," 3rd Edition, p. 418. *Dumb-bells of Oxalate of Lime.* It should be stated that a certain proportion of animal matter is deposited with the earthy or

crystalline material of which the globule is composed. This organic matrix may often be demonstrated when the highly refracting matter has been dissolved out by a chemical reagent.

Corpora Amylacea.—These are oval or circular masses, much resembling starch globules in their general appearance, which are found principally in the brain and spinal cord in disease, but they have been met with in many other fluids and tissues of the body, pl. XXV, fig. 18. Such bodies have been found in all parts of the organism in disease. They are very frequently associated with globules composed of phosphate and carbonate of lime. Virchow, in 1854, showed that they differed from the latter in chemical characters, and especially in becoming blue or greenish when acted upon by iodine.* Mr. Busk considers that some of these bodies are absolutely identical with starch.† I have described some which existed in considerable number in a cancerous liver, and could scarcely be distinguished from starch globules. *See* my "Lectures at the College of Physicians," 1861. Most "amyloid" bodies contain a large proportion of albuminous matter, and only a trace of the substance which exhibits the iodine reaction. The best iodine solution for testing their amyloid constituent is the chloride of zinc solution, the composition of which is given in § 198. Of the mode of formation of corpora amylacea, and of the consequences of their formation, but little certain is known.

242. Fibres, like *granules* and *globules*, may be formed independently of life. *A fibrous appearance* may be produced by drawing out any non-living viscid material in one direction. Under the microscope numerous lines are seen, the appearance not being due to individual fibres which can be isolated from one another, but to the linear stretching of the viscid material. Silica is an inorganic substance which in some states exhibits a fibrous arrangement very like that seen in many organic tissues. But definite fibres of extreme minuteness may be artificially prepared without difficulty. Glass may be drawn out into microscopic filaments as fine as the organic fibres of silk.

In the tissues of animals, besides true fibres, appearances are common which are due to the manner in which the tissue has, so to say, been laid down or deposited, and not to the development and formation of actual fibres. The subject under consideration will be further discussed after *elementary parts*, which used to be called "cells," have been referred to. For it will be shown that in many tissues "fibres" often bear to the formative bioplasm a relation corresponding

* Virchow's "Archiv." Band VI, s. 125.

† "Quarterly Journal of Microscopical Science," vol. ii, page 106. The following references may also be given on *corpora amylacea*. Dr. Carter, "Edinburgh Medical Journal," August, 1855, and "Graduation Thesis," 1856. Dr. Arlidge, "Medico-Chirurgical Review" vol. xiv, page 470.

to that which that constituent of the "cell," known as its cell wall, bears to the bioplasm concerned in its production.

Of the Bioplasm or Living Matter of the Body, and of Elementary Parts or Cells.

As the term "cell" is still constantly used, and the old "cell theory," in a modified form, continues to be taught by many lecturers and by most authors of text books, I am still constrained to treat the subject in this book in a manner that I feel is not quite satisfactory. If I could simply describe the structure that may be demonstrated after death, and the phenomena that may be observed in living elementary parts, my task would be easy, and the account I should give would be brief, and it would be easily understood, but as I am bound not only to describe the facts as they are known to me, but to make clear to the reader the particular points in which my views differ from those generally entertained, I must to some extent discuss the doctrines entertained by others, and show in what respects my conclusions differ from those arrived at by previous and contemporary observers.

243. Bioplasm and Bioplasts, Elementary Parts or Cells.—Many of the lower organisms throughout life, and all organisms, vegetable as well as animal, in the earliest stage of development, consist of a minute mass of clear transparent structureless living matter, possessing formative power of the most remarkable kind. This is *bioplasm*, or living, or *germinal* matter. When the *formation of tissue* occurs, as development advances, the outer part of a mass of bioplasm undergoes change, or—to express the fact in another way—part of the bioplasm dies, and a *fibre*, or *cell-wall*, or other matter having structure, or being structureless, results. This is *formed material*. These facts were demonstrated in my lectures before the Royal College of Physicians in the early part of 1861. An attempt has been made by more than one writer to unfairly prejudice students against my observations and conclusions by the suggestion that I have simply called matter already known as "Protoplasm" by another name. Now the writers in question must be aware that under the term *protoplasm* have been included, by various writers and by themselves, many kinds of *formed material*, each kind differing essentially from the matter I have called *bioplasm*, or *living matter*. As late as 1869, we find Mr. Huxley calling white of egg, dead muscle, and roasted muscle, and other things differing absolutely from every form of living matter, *protoplasm*. To this very day forms of non-living matter, as well as living matter, are termed protoplasm. But what makes the matter worse is the fact that the memoirs of Kühne and Max Schultze, which are often appealed to, and notably by one authority, in justification of the assertion, that my living matter is their protoplasm, were not published till

some time after my researches had appeared, in fact, not until after my volume, published in January, 1862, had been translated into German by Professor Victor Carus, of Leipzig (1862). The date of publication of Max Schultze's Memoir is 1863, and that of Kühne 1864!

The "cell" used to be described as a perfectly closed sac within which were certain *contents*. It was supposed that in its formation little particles became aggregated together to form collections, and that then the "cell-wall" was formed around these. It was afterwards maintained that the most important structure within the cell-wall was the *nucleus*, and to it was attributed the process of multiplication. Other important changes taking place during the life of the cell were referred to some mysterious action of the nucleus. But, on the other hand, it was affirmed by new authority, that in the *matter between the cell-wall and the nucleus* all those wonderful phenomena resulting in the production of characteristic tissue or cell products, occurred. Now it is to be remarked that the "nucleus" is not constantly present in all cells which, nevertheless, divide and subdivide, and that in many instances no distinction into *wall*, *contents*, and *nucleus* can be made. The human blood corpuscles exhibited no nucleus, and in order to explain the fact, and ensure these bodies being included in the cell-category, some authorities invented the hypothesis, that the red corpuscle was a "free" nucleus, while others insisted that it was a "cell," the nucleus of which had been absorbed.

In the "nucleus" of most cells a bright spot which could not be distinguished from an oil-globule was often observed. This was called the "nucleolus," and *nucleoli* generally, were regarded as nothing more important than oil-globules. An oil-globule, however, is a mere result of change; it is inanimate, incapable of growth and formation; but a true "nucleolus," where it exists, is *living bioplasm*, and is in fact *a new centre of growth.*

The cell "contents" are various. These differ no less in their physical characters than in their chemical properties and endowments, but every kind of *cell contents* was formed from *bioplasm.*

The elementary part itself may be destined to perform offices of the most temporary character, and its development, growth, and decay, may be comprised in an exceedingly short space of time, or the material of which the greater part of it consists may not be prone to alteration, the structure retaining its primitive "cell-form" unchanged for ages.

Elementary parts or "cells" cannot in all tissues be isolated or separated from each other, or from the so-called "intercellular substance" in which it is said certain forms of cells lie. They do not always appear as separate and individual structures, neither in many cases can any anatomical distinction be made between the cell-wall and

the so-called intercellular substance which intervenes between contiguous cells.

The "cavity" of the cell has been regarded as a little space, scooped out as it were in the material, of which the so called cell-wall or intercellular substance consists. The cavities it has been said result from "differentiation" taking place in a previously homogeneous plasma, but this cannot be, for the "cavities" contain the living matter which existed before the surrounding matrix was formed.

Endoplast and Periplast.—To the entire contents of the "cavity" or cell (where they can be removed as an independent mass), Professor Huxley gave the name of *endoplast*, and to the walls of the cavity, or the cell-wall and the intervening material or basis substance, that of *periplast* (1853). This observer erroneously considered the *periplastic substance* to be the formative matter, and believed that it alone took part in the differentiation which resulted in the formation of tissue. The *endoplast*, on the other hand, was regarded by him as a substance of comparatively little importance, and he even went so far as to assert that it was sometimes absent, and that its presence might be considered accidental. Huxley did not, however, state how he proved the truth of his extraordinary doctrine, nor did he tell his readers whether the endoplast was absent in a cell actually growing and changing. His statements are opposed to the fact that "endoplasts" are or have been wherever periplastic substance is found, while they often exist although the latter is absent. "Endoplasts" are far more constant and numerous than used to be supposed. They are present in all tissues, and numbers which have entirely escaped observation and cannot be seen if the ordinary methods of investigation are followed, may be demonstrated readily in the very same textures by special methods of enquiry. See page 64.

With reference to Huxley's theory, I need only remark:—first, that as there is no instance known in which any form of tissue is produced without the matter termed by him *endoplastic* being present, we are not justified in inferring that this is non-essential, but on the contrary, we are rather led to the conclusion that it is of the highest importance; secondly, that in the natural growing state, the periplastic substance is continuous with the endoplast. I have shown that the former is formed by the latter—in fact, that the *endoplastic* gradually undergoes change and becomes converted into the *periplastic* matter; and must therefore be considered as *primary and essential;* and lastly, that while periplastic matter cannot produce more periplastic matter like itself, the endoplastic substance can not only give rise to more matter like itself, but in the process of formation becomes resolved into periplastic substance. It seems then that the endoplastic matter must be of higher importance than that which surrounds it and which is formed by it. It unquestionably

exists before the periplastic substance is formed, it exists whenever the latter is being produced, it only is active, capable of increase, of growth, of formation, and yet Huxley says it is of no importance.

244. Of the real Structure of Elementary Parts or Cells.—The confusion resulting from the different views advanced, and the conflicting statements of different authorities, rendered it imperative to re-open the question of cell formation. In the lectures which I delivered at the Royal College of Physicians, in 1861, I entered fully into the question, and I think the matter may now be brought under the notice of the student in a much clearer form if I approach it from a point of view somewhat different to that usually taken. Instead of drawing conclusions from the structure of "cells" in fully formed tissues, let us examine "cells," or the bodies corresponding to them at different stages of growth, commencing at the earliest period of their existence. If we examine any embryonic matter, or any other rapidly growing material, we shall find that it is made up of small masses of transparent semifluid matter exhibiting no structure, but manifesting remarkable powers or properties. Each one of these masses is capable of moving, of dividing and subdividing, and of undergoing conversion into matter which did not exist before. From its transparency, this matter is often passed over when embedded in tissue, but it exists even in bone and teeth, at least whenever the formation of new bone or tooth structure is going on, and its presence can always be detected by the use of the carmine fluid, p. 65. When a number of particles of such matter are seen together, the mass has been said to consist of free "nuclei," "nuclear corpuscles," or to be composed of "granular matter." The actual "granular matter" entering into the formation of the living substance under examination is, in fact but the debris resulting from change occurring after death, for oftentimes the examination is not made until the living matter has long been dead and has undergone disintegration and decomposition. The living matter of the cell undergoes great alteration very soon after death. In its natural living, growing state, it may be so transparent and clear as to be passed over entirely, but when death occurs, oil globules and earthy particles may be deposited, and these among other things give rise to the "granular appearance." Frequently the living matter shrinks soon after death so that it occupies much less space than when it was in its living state, and instead of being continuous with the formed material, as was the case during life, becomes separated from it by an interval. This is often seen in cartilage, and the change which has taken place has led many to the erroneous conclusion that in that tissue a "cell," "granular corpuscle," or "endoplast," lay free in a cavity which existed in the cartilage matrix. The true relation of the living matter to the formed matter can be readily demonstrated by the student in perfectly

fresh embryonic cartilage, especially that of the young newt or frog tadpole, in which the anatomical elements are very large and easily discerned in a very thin section.

The student may form a good idea of living, growing, active matter if he examines a white blood corpuscle, a mucus corpuscle, or a pus corpuscle (pl. XXVI, figs. 7 to 11), under a power of 500 diameters, and upwards. Under still higher magnifying powers, he may see particles of the same active matter far more minutely. It is this matter which is alone concerned in the production of everything that lives. It was derived from matter which existed before it. All germination, all growth and multiplication depend upon it, and as every kind while in a living state possesses the power of germinating, of giving origin to matter like itself, I have termed it *germinal* or *living matter* or *bioplasm.* This living matter exhibits phenomena not known to occur in any non-living matter whatever, and therefore the term *living* as I employ it, has a very definite signification. Although we do not know the nature of the changes we call *vital*, we do know that they essentially differ from any chemical or physical changes yet discovered.

A free mass of bioplasm soon becomes changed upon its external surface. Surrounding conditions alter it, and this part soon loses its power of germination, and ceases to be active and living. The matter formed in consequence of the changes which occur, protects that which still remains, and prevents it from undergoing the same alteration or only permits such a change to go on very slowly. As this last passive matter is formed from the first, I have called it *formed material.*

Every elementary part or cell consists of living active *bioplasm*, surrounded by passive *formed material.* This layer of *formed material* may be so soft and diffluent, as not to form a layer to which the term envelope or wall could be correctly applied,—and so transparent and structureless as to elude ordinary observation ; or it may be so firm and thick, and may exhibit such remarkable structure and be present in such large quantity in proportion to the bioplasm, that the "cell" appears to be entirely composed of it. This is well seen in the hard cells of which the shell of nuts is composed.

Different kinds of bioplasm or living matter possess different power of resisting the influence of external conditions. Some kinds are easily killed —others are destroyed with difficulty. In many instances the vitality of bioplasm is only retained at a certain temperature, a difference of a very few degrees being sufficient to destroy its life. Some forms resist extreme cold, but as far as is known none can live at a temperature above 300°.

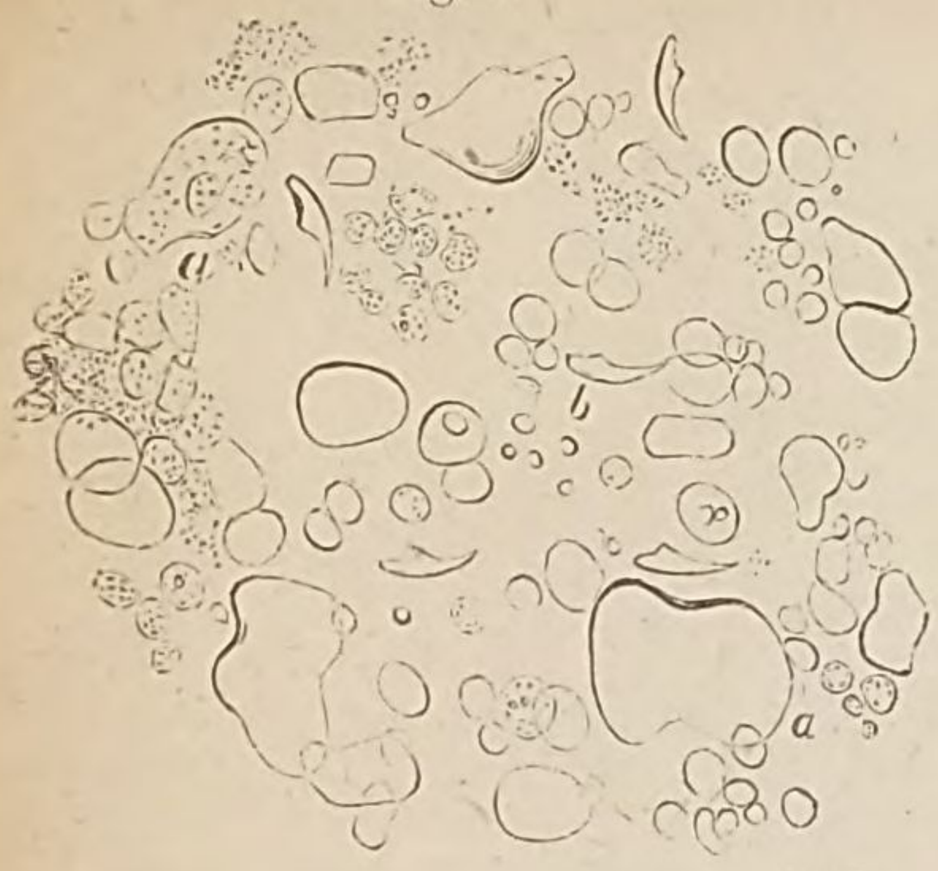
Some kinds of living matter may pass long distances through air or water without being destroyed. Sometimes they multiply during their transit, but sometimes remain quiescent, it may be, for a long period, multiplying only when they happen to fall upon a surface where the

particular pabulum adapted for their nutrition, and circumstances favourable to their development, are present. It is probable that the *materies morbi* or *virus* of all contagious diseases consists of living bioplasm capable of retaining its vitality in spite of the influence of many adverse conditions, and even when dry. Desiccation, however, is not complete—a little of the living matter being protected by the covering of formed material, and retaining sufficient moisture to keep it alive. Thus, living particles of vaccine lymph, of the virus of small-pox, of scarlet fever, and of many other diseases, may retain their vitality although apparently dried up, just as many of the infusoria and lower vegetable organisms may live quiescent in a state of imperfect desiccation for a length of time,—growing and multiplying rapidly as soon as favourable external conditions become established. See p. 126, pl. XII, figs. 3, 4, 6, showing the bioplasm of pus and vaccine lymph. These particles of bioplasm are not then to be regarded as low animal or vegetable forms, but as the direct but degraded descendants of the bioplasm of the normal tissues of the organism. Just as under certain conditions the bioplasm, known as pus, results from that of normal cells,—so, under other conditions not yet perfectly understood, the living particles inducing specific contagious maladies, and capable of multiplying in the blood of one person after another, originate from normal bioplasm.

It must not be supposed that we can form any conception of the *power* of the different forms of bioplasm from microscopical or chemical examination. Masses which could not be distinguished from one another, manifest the most remarkable differences in power. For instance, fig. 9, pl. XXVI, much resembles fig. 10, but the first represents bioplasts formed during ordinary inflammation tending to the production of the low form known as *pus*, while the last represents the highest form of bioplasm in nature—the brain cells of man at an early period of development. By chemical analysis every kind of bioplasm yields—1, a substance resembling *fibrin*; 2, another allied to *albumen*; 3, *fatty matter*; 4, *salts*; and 5, *water*.

Every "cell" in nature consists of *bioplasm* or *living matter*, and *formed material* or passive or dead matter produced from the first, fig. 1, pl. XXVII. The proportion and characters of the formed material vary infinitely. Elementary parts or cells differ widely in characters, property, and actions, but every one is capable of growth and change, and may be proved to consist at every period of its life, of bioplasm and one or several kinds of *formed material*. However different in their fully formed state elementary parts may be, every one of them at an early period of its formation consists of bioplasm exhibiting precisely the same fundamental characters, as far as we are able to determine at present. There are no known tests or methods of examination, by which the bioplasm of any animal could be distinguished from that of man.

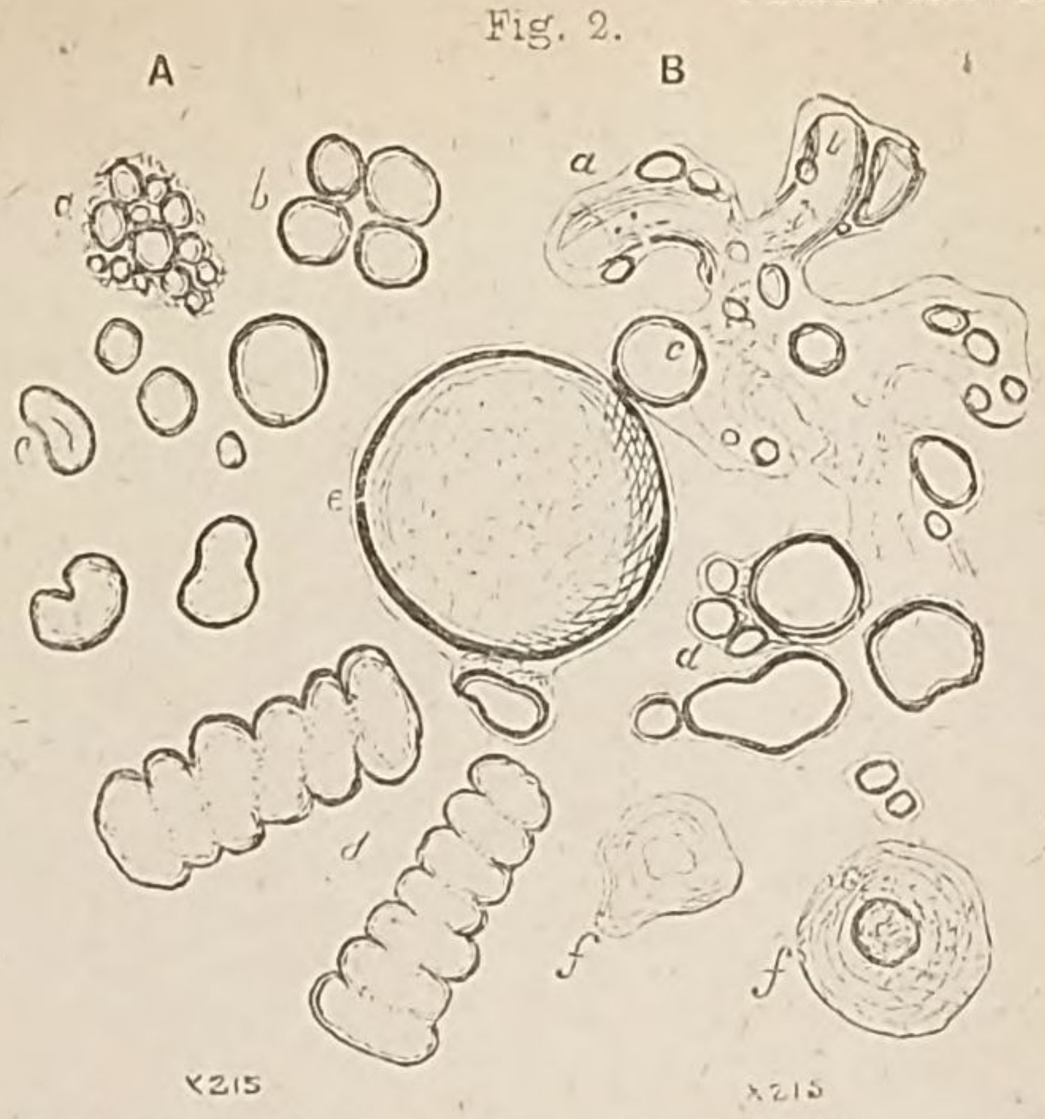
Fig. 1.



×215

Deposit from fluid removed from the chest of a girl æt. 23. *a*, granular cells, probably from the walls of the cavity in which the fluid had collected *b*, claws of echinococci. The circular oil-like masses were rendered granular by acetic acid. p. 223.

Fig. 2.



×215 ×215

Fig. 2.—A, globules of earthy matter, cerebellum. Case of insanity. Consisting principally of carbonate of lime. *a*, collection of small globules. *b*, larger ones. *c*, globule resembling corpus amylaceum. *d*, collections of globules incorporated together. × 215. B, globules of earthy matter. Choroid plexus. Insanity. These were soluble in hydrochloric acid without effervescence, and consisted almost entirely of phosphate of lime. *a*, capillaries of one of the processes of the plexus, showing the globule external to the capillary wall. *b*, outline of a capillary loop. *c*, large globule external to capillary. *d*, small collections enclosed in a membranous structure. *f*, globules acted upon by hydrochloric acid. × 215. p. 223

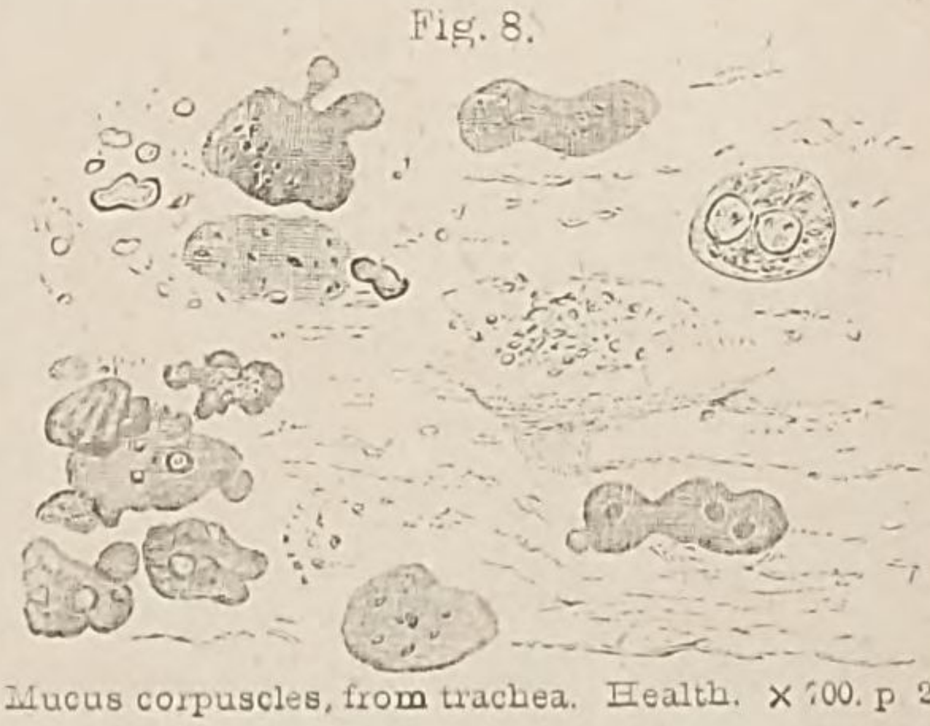
Fig. 3. Fig. 4. Fig. 5. Fig. 6.

The appearance of crystals of carbonate of lime, from horse's urine, as examined in different media. 3, by reflected light in air; 4, by transmitted light in air; 5, in water; 6, in Canada balsam. p. 223.

Fig. 7.

White blood corpuscle, showing transparent substance, which forms protusions or outgrowths. Granules (Bacteria genus?) embedded in the outer oldest portion of this; also the large nuclei. × 2800. pp. 231, 258.

Fig. 8.

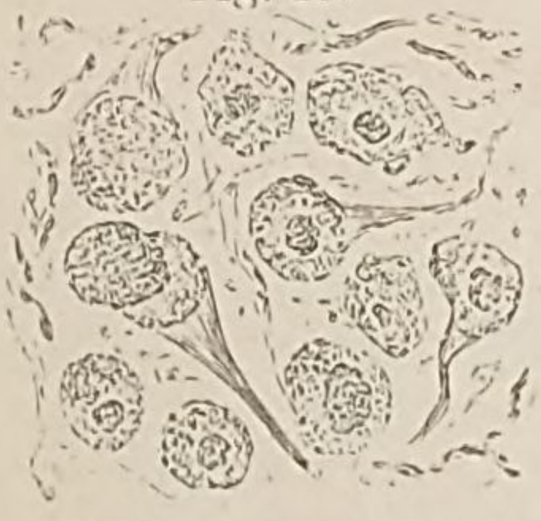


Mucus corpuscles, from trachea. Health. × 700. p 229.

Fig. 9.

Lymph on surface of peritoneum of intestine Acute peritonitis 4th day. × 700. p. 230

Fig. 10.



Anterior portion of cerebrum. Human embryo. 4th week. × 700 p. 230.

Fig. 11.

One of the mucus corpuscles like those in Fig. 8, magnified 2800. Showing alterations in form during a minute. p. 229.

[To face page 230.

245. Formation of Elementary Parts or Cells.—The mode of production of some of the principal forms of elementary parts or cells in the adult state, may perhaps be understood from the following outline, with the aid of the drawings in the plates. It is only by studying the changes through which the cell passes up to the time when it acquires its permanent type, that the student can gain a correct knowledge of forms, or obtain a clear conception of the wonderful process of the development and formation of tissues and organs.

I. The mass of bioplasm may absorb nutrient material from the surrounding medium, increase in size, divide and subdivide according to the process described on page 137. Thus one mass may give origin to many, which grow into masses like the first. In such case, as the one under consideration, very little formed matter is produced, and this may be fluid, semifluid, or soft and yielding, so that the masses of bioplasm easily move, and divide, and subdivide in it, pl. XXVI, figs. 8 and 11. The entire quantity of living matter or bioplasm increases rapidly and at a much greater rate than the formed material. Bioplasm divides, and the masses multiply in this way during the early period of development of all textures, and in all rapid growth occurring in disease. In inflammation and in cancer the rapid growth of bioplasm is remarkable.

II. A mass of bioplasm may undergo change upon its external surface so that an envelope of formed material is produced. This may be thickened by the production of more formed material which is invariably deposited layer after layer within *that first formed*, pl. XXVII, fig. 1, 2, 3, 4.

III. A *thick layer* of formed material having been produced, the living matter or bioplasm within may die, or may be the seat of some of the other changes mentioned below.

a. The bioplasm may undergo very slow change, and remain as a small mass embedded in a great quantity of formed material.

b. The bioplasm may remain for a long time in a state of comparative quiescence within this protective covering, but if exposed to favourable conditions the formed material becomes softened, and the bioplasm makes its way through it at certain points, growing rapidly when it comes into close relation with the pabulum in medium around, fig. 7, *a*, pl. XXVII. In the case of the vegetable spore of mildew for example, the appearance represented in fig. 8 results. The bioplasm having occupied all the space left white in the drawing.

c. The small remaining particle of bioplasm may die, in which case a mass consisting entirely of *formed material* results. A very small cavity remains in the centre which is due to the absorption of the materials resulting from the death of the bioplasm. The whole constitutes a mass of dead matter, and is incapable of formation, conversion or multiplication. As an instance, the outer-

most and oldest "cells" of the cuticle, or of a mucous membrane may be adduced, fig. 2, *e*, pl. XXVII.

d. The bioplasm of an epithelial cell, as represented in *c*, figs. 1, 2, remaining alive, may increase again in size at the expense of the formed matter it has already produced. Such changes are common enough in inflammation, and will be at once understood by reference to plate XXXIII, p. 250, fig. 1, *a*, *b*. At last the whole of the formed material is appropriated by the bioplasm, and the appearance represented at *f* and *g*, fig. 2, results; in fact, a return to the state of things observed at an early period of development. But although little or no difference can be discovered by microscopical or other methods of examination between masses of embryonic bioplasm and those resulting from the increased supply of pabulum which reaches the bioplasm in the pathological process of inflammation, there is remarkable degradation in the power of the rapidly-growing masses of bioplasm. They can never again form any lasting texture, or give rise to anything exhibiting definite structure or capable of performing any special office.

IV. A mass of bioplasm having undergone conversion into formed material upon the surface, so that a permeable membrane or cell-wall is produced, may also give rise to the formation of a peculiar material within. In the substance of the bioplasm, a little particle, resulting like all formed material from the death of bioplasm, makes its appearance, and gradually increases in size by the addition of new matter to it of the same kind. In this way fatty and starchy matters are formed, as is said, *within* the cell. But these, like the external membrane or cell-wall, consist of formed material, and result from the death and change under certain conditions of particles of the bioplasm. The mode of production of starch will be understood by reference to figs. 9, 10, pl. XXVII, and that of fat by referring to figs. 1, 2, pl. XXVIII. In some cases the matter, instead of taking the form of distinct grains or particles in the interior of the cells, is added layer after layer upon the inner surface of its walls. But the deposition of this "secondary deposit" is not uniform. Nutrient matter is continually passing towards the bioplasm, and deposition will occur in the intervals between the lines in which the nutrient juices flow. At last a star-like appearance results. The manner in which this occurs will be understood by reference to fig. 3. The lacuna of bone is formed in somewhat the same way.

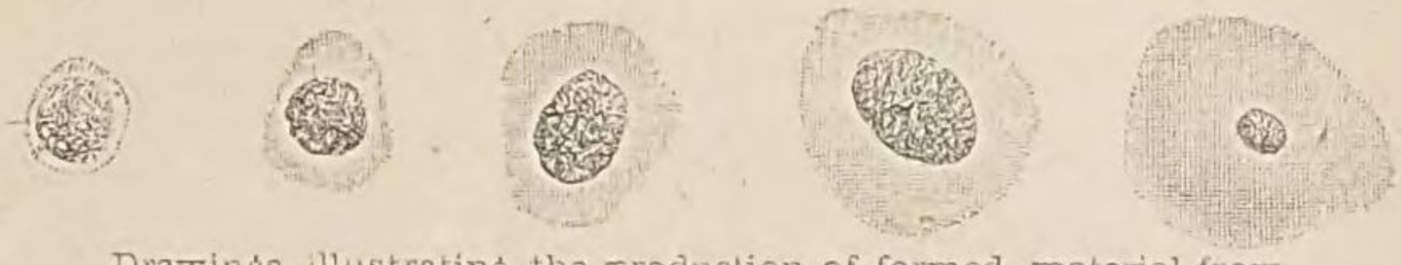
V. A mass of bioplasm, lying a short distance external to a vessel, and having given rise to the formation of a certain amount of formed material, may continue for a long time to effect most important changes, although the mass itself appears not to change. Having one surface near to, or directly in contact with, the vascular wall, while the other

PLATE XXVII.

Fig. 1.

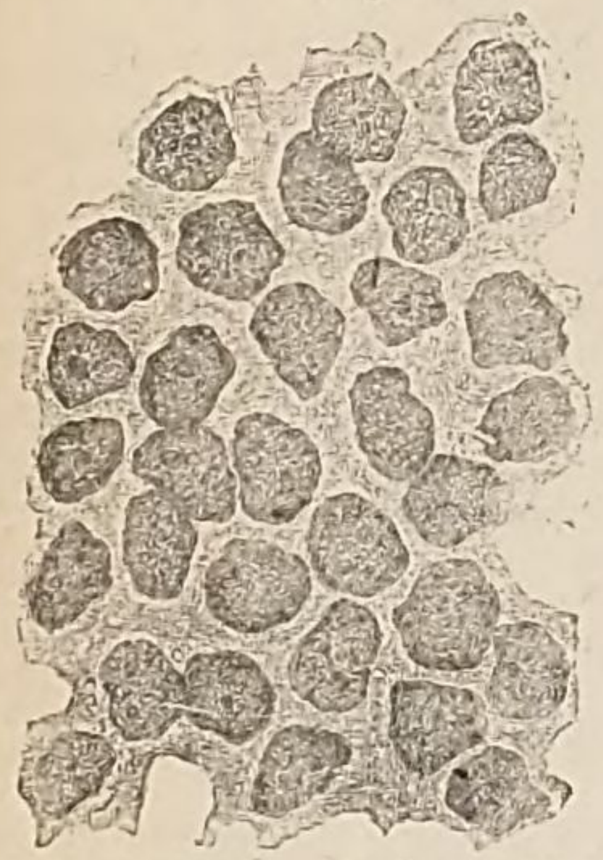
Production and accumulation of formed material upon the surface of bioplasm in an epithelial cell, as in cuticle. × 700 p. 230.

Fig. 2.



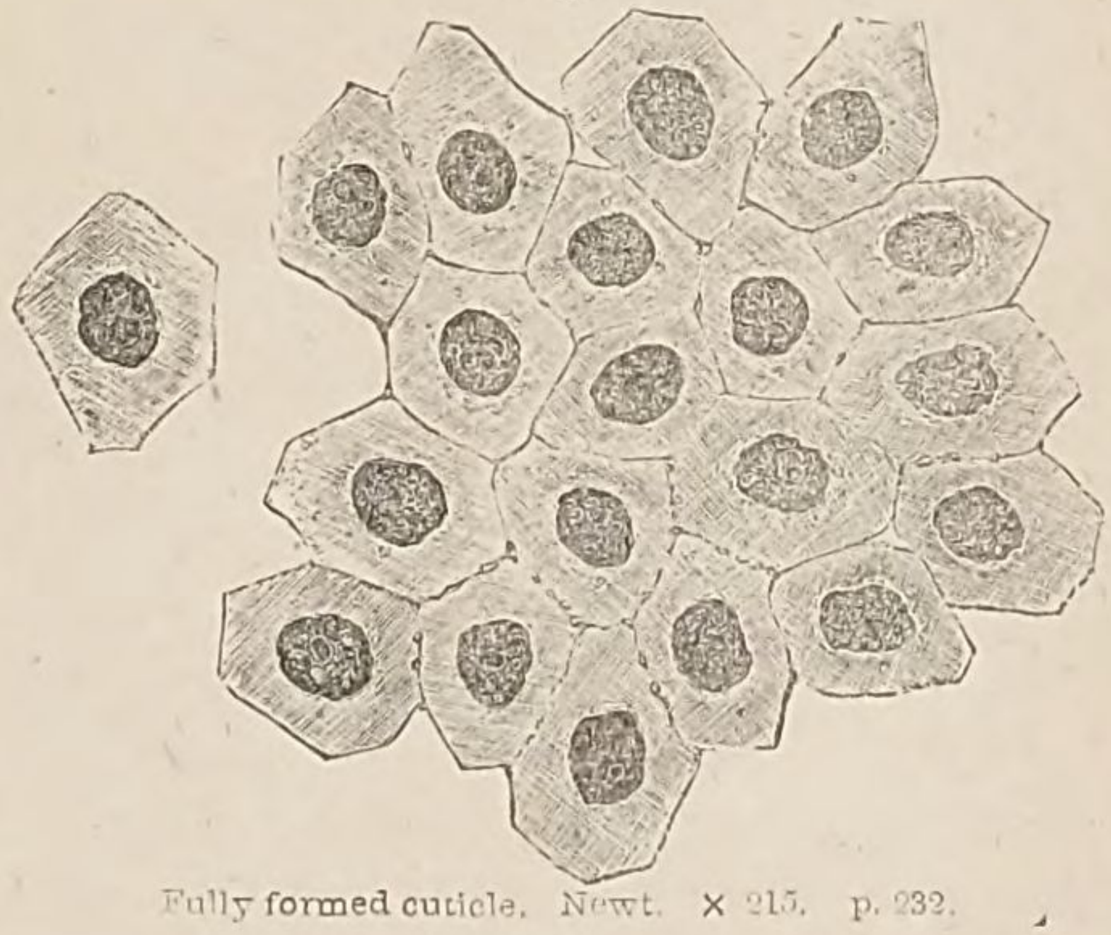
Drawings illustrating the production of formed material from the bioplasm in epithelial cells. p. 232.

Fig. 3.



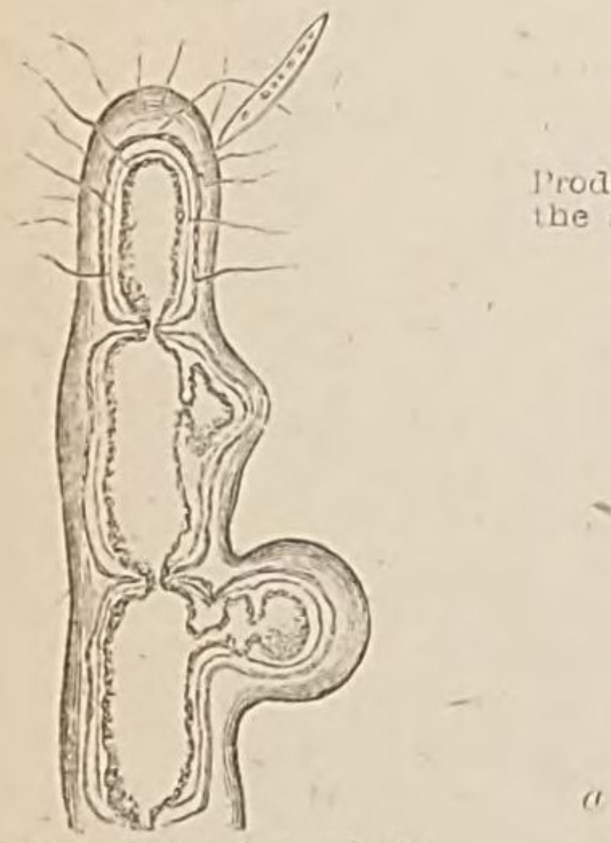
Young cuticle. Newt. × 215

Fig. 4.



Fully formed cuticle. Newt. × 215. p. 232.

Fig. 5.

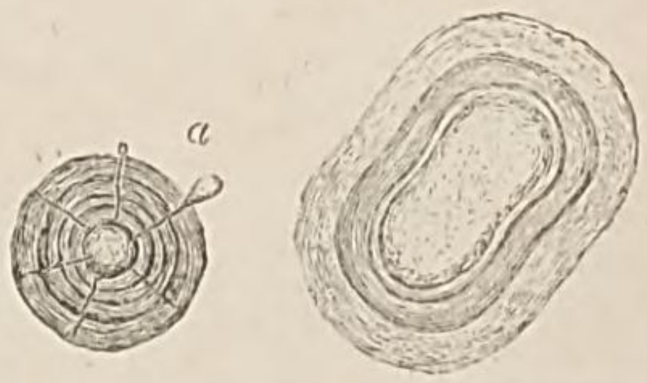


Growing extremity of the stem of a sea-weed, showing the manner in which the bioplasm divides, and the way in which the formed material is deposited upon its surface, layer within layer. The outermost layer is the oldest, and is undergoing disintegration. Other organisms are living upon it. × 350. p. 231.

Fig. 6.

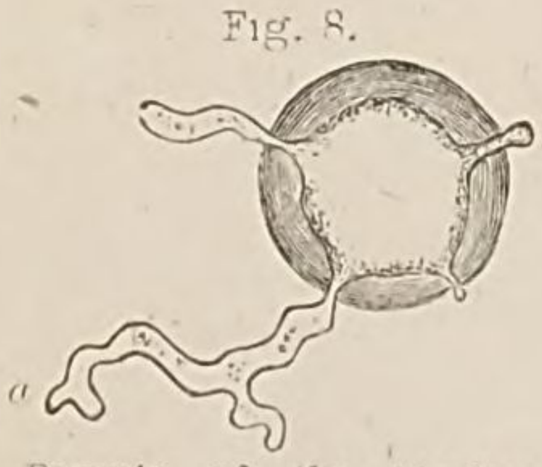
Production of formed material upon the surface of bioplasm of mildew. × 1800. p 229.

Fig. 7.



Further production of formed material in ordinary mildew. At *a*, a bud is formed by the passage of some of the bioplasm through pores in the very thick layer of formed material. × 1800. p 231.

Fig. 8.

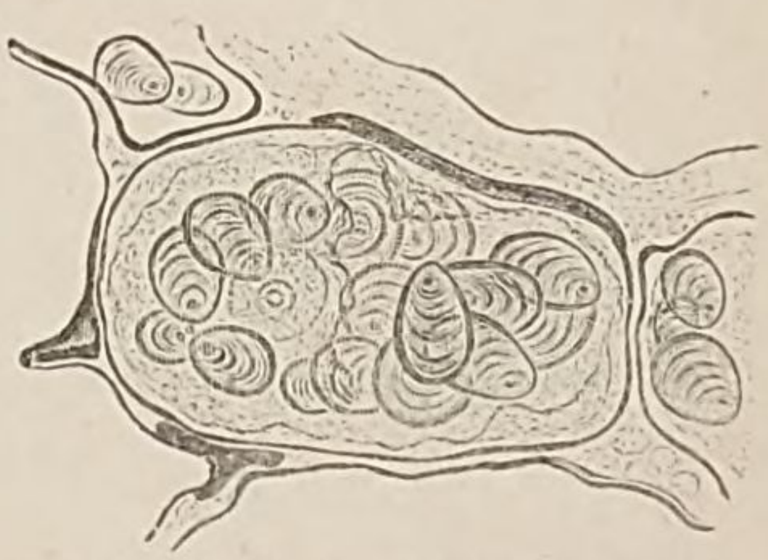


Passage of the bioplasm through pores in formed material, and formation of thin layer of formed material upon bioplasm. *a*. Showing the manner in which fungi grow × 1800 p 231.

Fig. 9.

Five young starch holding cells of the potatoe, showing bioplasm, with small starch globules precipitated amongst it. × 700. p. 232.

Fig. 10.



A fully formed starch holding cell of the potatoe. × 350. p 232.

1000th of an inch ——— × 700.

[To face page 232.

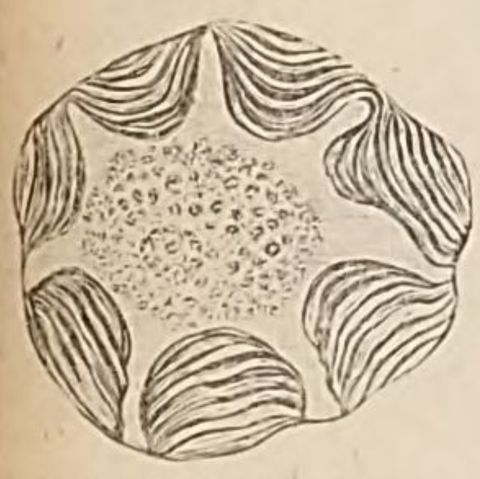
Fig. 1.

'Fat Cells,' showing the seat of formation of the fat (formed material), and the changes occurring in the cell as it advances towards its fully developed state. Frog. × 700. p. 232.

Fig. 2.

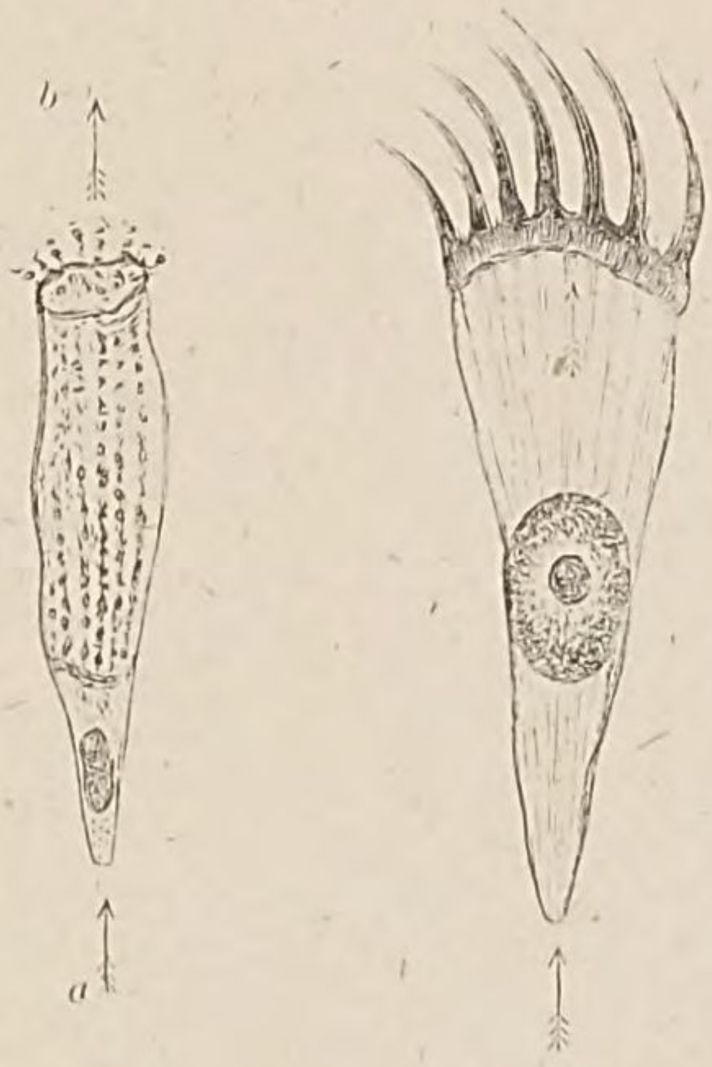
Adipose tissue, abdominal cavity. Newt. The nuclei of many of the fat vesicles are unusually distinct. × 215. p. 232.

Fig. 3.



Vegetable cell, showing the manner in which secondary deposits are formed, and how the channels through which currents flow towards the nucleus' result.

Fig. 4.

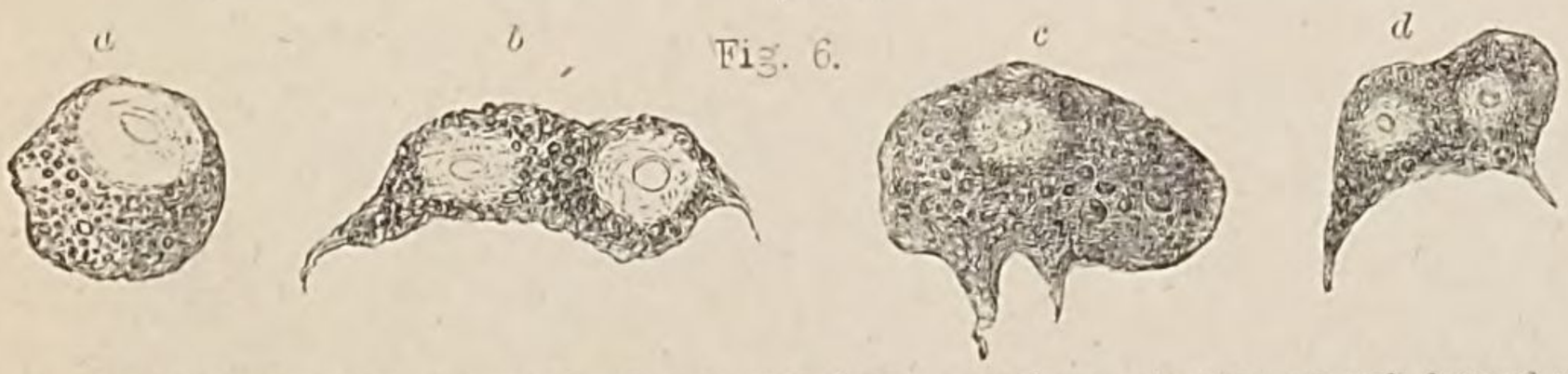


Mucus-forming and Ciliated Cells, from the fauces of a boa, showing bioplasm and the formed material within the envelope. The lower arrow, *a*, shows the direction in which the nutrient pabulum flows towards the bioplasm, the upper one, *b*, that taken by the formed material as it passes from the bioplasm. p. 238.

Fig. 5.

Columnar Epithelial Cells, from the small intestine of a dog, showing the position of the bioplasm. *a*, the layer of highly refracting matter upon the surface. × 700. p. 237.

Fig. 6.

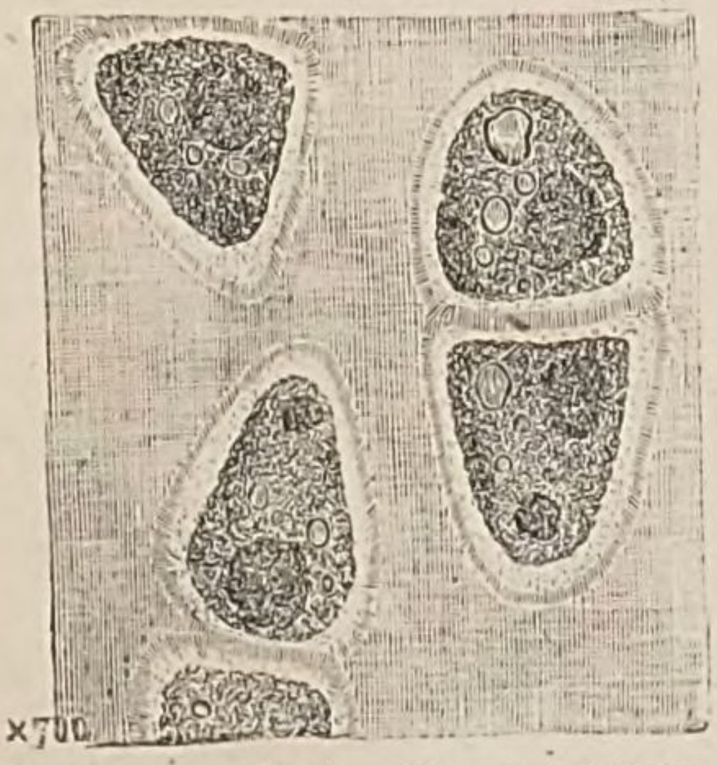


Liver Cells in different stages of growth, showing bioplasm and the production of soft formed material, which becomes resolved into several different substances (fatty, amyloid, albuminous, and biliary matters.) Human subject. × 700. pp. 233, 235.

Fig. 7.

To show how the appearance of separate cells is produced in formed material. p. 233.

Fig. 8.



Cartilage. Frog. Showing bioplasm and formed material (intercellular substance, of authors), with appearances resembling a cell-wall. × 700. p. 233.

Fig. 9.

To show how in some tissues the formed material is continuous. p. 233.

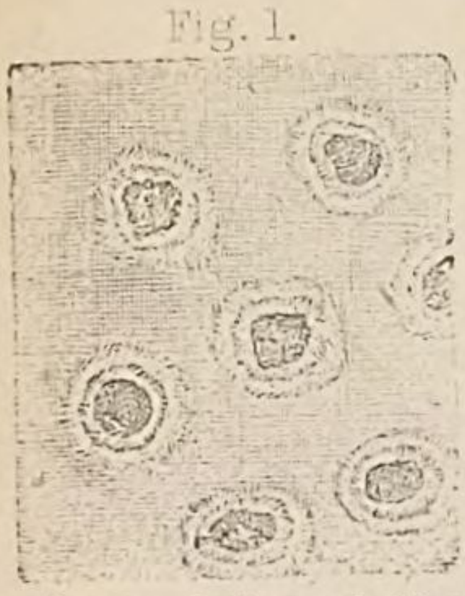
1000th of an inch | | × 215.

" " |_____| × 700.

[To follow Plate XXVII.

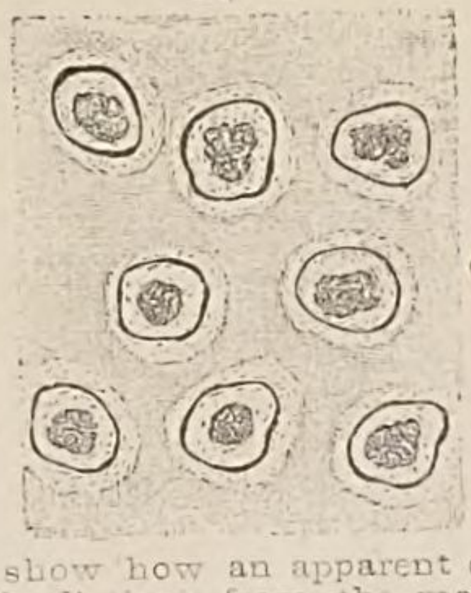
PLATE XXIX.

Fig. 1.



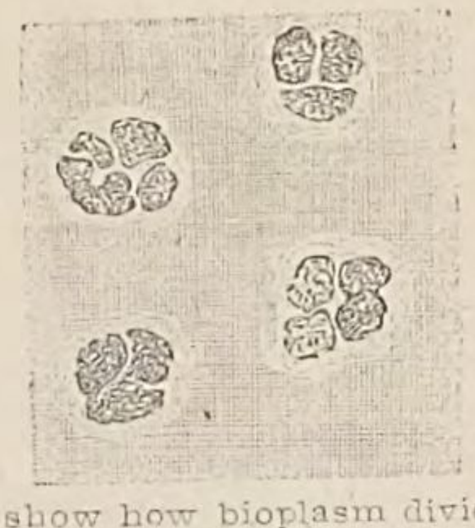
To show relation of bioplasm to formed material of cartilage p. 233.

Fig. 2.



To show how an apparent cell-wall, distinct from the matrix may result. p. 233.

Fig. 3.



To show how bioplasm divides to form collections of cells separated by matrix. p. 233.

Fig. 4.

Tendon. Young. × 215.

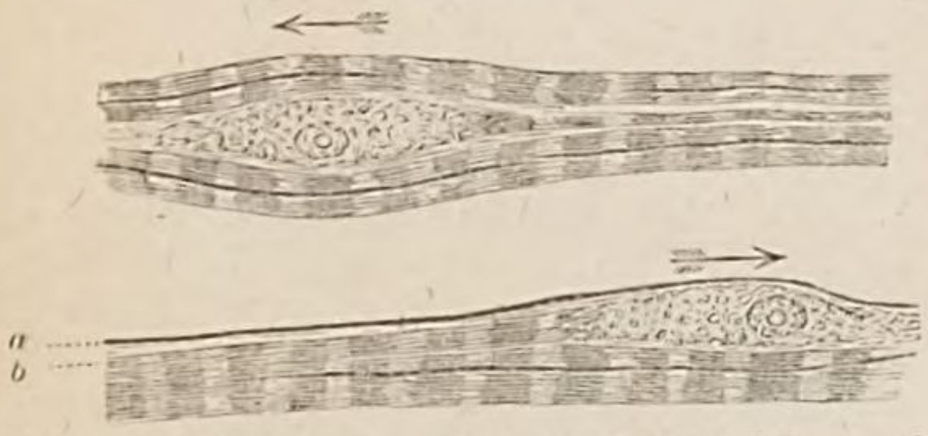
Tendon. Fully formed. × 215.

Showing bioplasm and formed material (intercellular substance, of authors) of tendon at different stages of development. The granular masses indicate the position of the bioplasm. p. 233.

Fig. 5.

Stellate tissue on surface of fang of a human incisor tooth. × 700. p. 231.

Fig. 6.



Muscle. Bioplasm and formed material. *a*, the sarcolemma. *b*, the contractile material. The arrows show the direction in which the masses of bioplasm are supposed to be moving. p. 233.

Fig. 7.

Portion of a fibre of yellow elastic tissue. Ligamentum nuchæ, Lamb. The mass of bioplasm, *a*, is moving in the direction of the arrow, behind it the formation of yellow elastic tissue is proceeding. p. 233.

Fig. 8.

Vaginal epithelium from urine. × 215. p. 237.

Fig. 9.

Bladder epithelium. *a*, from the general surface. *b*, from the fundus. *c*, scaly epithelium from the bladder. × 215. p. 237.

1000th of an inch |___| × 215.

" " |_____; × 700.

[To follow Plate XXVIII.

is free in the cavity of the vessel or in that of a gland follicle or cell,—nutrient matter may be absorbed by the bioplasm on one side, while on the other the production of formed material in the form of solid, liquid, or gas proceeds. This may become resolved by oxidation or other changes into matters which escape from the free surface and are readily carried away. Such are the phenomena which occur in the processes of absorption of nutriment and in secretion. Different forms of secreting cells are represented in fig. 4, 5, 6, pl. XXVIII.

VI. A mass of bioplasm may divide and subdivide into several masses. The intervening formed material, instead of being arranged round each mass, as in pl. XXVIII, fig. 7, may be continuous throughout, as in fig. 9. After a time the formed material may undergo condensation, but the shrinking that would take place may be compensated for, or more than compensated for, by the continual formation of new formed material. A tissue like cartilage results, fig. 8.

The formed material may be uniform, transparent, or granular, or it may exhibit a fibrous appearance, according to the conditions present during its production or subsequently, and the movements of the masses of bioplasm which produce it. It has been assumed by many, amongst whom must be cited the distinguished Kölliker, that the formed material of cartilage is deposited between the cells by a separate operation; and that it is therefore truly, *intercellular substance.* If this were so the cells would simply become separated from one another as the texture advanced in age, their cell-walls remaining distinct, while the changes in character which may be observed in them at different periods of the growth of the tissue would not be accounted for.

After a time each of the masses of bioplasm lying at intervals through the formed material of cartilage may again divide and subdivide, each of the masses resulting from subdivision giving rise to the production of formed material on its surface for a time, and then dividing and subdividing. There will now be found primary and secondary collections of masses, separated from one another by layers of formed material varying in thickness, fig. 3, pl. XXIX.

In some cases the surface of the mass of bioplasm is changed into a layer of formed material, in which case the appearance of a cell-wall distinct from the matrix results, pl. XXIX, fig. 1. Or, the bioplasm may shrink and become disconnected with the formed material which has already been produced by it. Becoming then hardened upon its surface, the appearance represented in fig. 2 results.

VII. Masses of bioplasm may separate from one another in a linear direction, and the formed material may accumulate as a thread between them. This is illustrated in the production of white fibrous tissue, yellow elastic tissue, and muscle, pl. XXIX, figs. 4, 6, 7. In some cases the oval mass of bioplasm may continue to move along the thread

or fibre already formed, and add to its thickness. In this way the thick fibres of yellow elastic tissue result, and in certain forms of muscle, particularly that of the heart and tongue, the fibrillæ are thus increased in thickness and new ones formed, figs. 6, 7. If the separation of the masses of bioplasm takes place in various directions, a tissue consisting of "stellate cells" will result, as is well seen in fig. 5.

246. Of the different kinds of Cells and Elementary Parts.—Elementary parts differ in their chemical and physical properties, and in the duration of their existence. They may be distinguished according to the offices they discharge, or they may be classified according to the peculiarities of form alone. The latter plan is generally adopted when varieties of cell formation are described; and *scaly* or *squamous* cells, *tessellated* cells, *polygonal* cells, *columnar* cells, *spherical* cells, *spindle-shaped* cells, *fusiform* cells, *fibre* cells, and *caudate* cells, some of which are very complex, have been distinguished. It would seem, however, a more natural system to arrange cells in groups according to the offices they discharge in the organism. Thus we should have cells whose office it is to form a protective covering to delicate structures placed beneath them; cells which are specially concerned in separating and elaborating certain materials derived from the blood, and which at length form the special constituents of different secretions; cells which give rise to currents in the fluid which bathes their surface, by the perpetual vibration of minute hair-like appendages or *cilia* developed on their free surface. Besides the above there are cells with special endowments such as *contractile cells* and *nerve cells;* cells whose nutritive changes are associated with the development of heat, light, or electricity, or the manifestation of other modes of force; cells taking part in the reception of external impressions, as those concerned in touch, taste, smell, hearing, sight; lastly, there exists an almost infinite variety of cells in different morbid growths, which differ essentially from the cells of healthy tissues, but which have nevertheless directly descended from the bioplasm of cells developed in the normal state.

All cells possess the power of mutiplying in number, of selecting and appropriating certain materials, and of rejecting others. So also they have their periods of growth, development, and decay, and the death of each takes place at its appointed time. Some seem destined to absorb large quantities of matter and pass it onwards into channels adapted to receive it. The power of multiplying with wonderful rapidity in the normal state seems peculiar to some, while the most striking character of others is their power of selecting and slowly converting certain substances into the constituents of the secretions. Nevertheless the bioplasm of all may, under certain circumstances, grow and multiply with great rapidity.

The *secreting cell*, pl. XXVIII, fig. 6, is distinguished by its more

or less spherical, or polyhedral form and soft granular formed material (cells of liver, kidney, pancreas, &c.) ; the cell concerned in *absorption*, by its columnar form and by its thickened and spongy outer extremity (columnar epithelium of intestine, ducts of salivary, pancreatic, labial, and buccal glands, liver, &c.). The cell which only serves the office of a protective covering to delicate structures beneath, is known by its hardness and density, by its flattened appearance and imbricated or tessellated arrangement (squamous epithelium of skin, mucous membrane of mouth, œsophagus, vagina, &c., tessellated epithelium covering the surfaces of serous membranes, &c.).

The *cell-wall* and *cell-cavity* are not to be demonstrated in all cases, and many structures which are still called *cells*, have been shown to consist of masses of material arranged in shapes like cells, but not invested with any membranous envelope. So also examples are not wanting in which granules, globules, and other matters have collected together, and gradually firm, compact, *cell-like* masses have been formed. In different specimens of sputum, small collections of dark granules are often found. In many cases these are, without doubt, mere aggregations of particles of carbon introduced into the air-tubes during inspiration. By the action of the currents produced by the vibration of the cilia, these become mixed with a little mucus, and at length are formed into nearly spherical bodies which exactly resemble cells. Not unfrequently, the mucus deposited on the exterior so closely resembles a cell-wall, that it is difficult to believe these granules are not really inclosed in a cellular envelope. The flattening and gradual extension, rather than rupture, which such masses undergo by pressure, the circumstance of their being found in all the different stages of increment, and the action of chemical reagents upon them, prove conclusively that they are formed in the manner I have described. Some forms of the so-called "granular corpuscles," "compound granular cells," or "inflammation globules," appear to be formed in the same way. It is not possible to demonstrate a cell-wall in many other cases, and I have proved conclusively that the *liver-cell* is destitute of any membranous wall.*

* "In a large number of animals, then, the contents of the tubular network may be said to be continuous ; in some it is interrupted so as to form masses irregular in size, in which nuclei are scattered at intervals ; and in others, the particles are more uniform in size, resemble each other very closely in general character, and each contains a separate nucleus. Between the numerous, well-defined, and separate masses, or liver-cells of the mammalian animal on the one hand, and the continuous mass which occupies the tubular network of the fish on the other, it is easy to demonstrate every intervening shade of difference ; and more than this, at different periods of development of the embryo, and in various morbid conditions of the human liver, every degree of separation and continuity may be observed. Again, by the action of various chemical reagents as described in page 40, the distinct and separate cells of the healthy mammalian liver may be made to fuse, as it were, so as to form continuous masses,

The fully-formed blood corpuscle is not a *living* cell. It seems to be a little mass of viscid matter the outer part of which is tolerably firm. It is permeable to fluids holding various solid and gaseous substances in solution, in both directions. It is oval or circular in form, and becomes bi-concave or bi-convex as the density of the medium in which it is immersed is altered. Fluids of high specific gravity, and oxygen, flatten the corpuscles, while water, fluids of low density, and carbonic acid, cause them to become swollen and more or less spherical. By allowing blood corpuscles to soak for some time in fluids of low specific gravity, it is said that they burst, and their contents escape, but really the matter of which the entire corpuscle consists, imbibes water, and gradually dissolves. The outer part of the matter of which the oldest corpuscle is composed, is, however, often hardened and presents the characters of a cell-wall.

It is not consistent with the plan of the present work, to describe in order the different structures met with in the human body, and I shall only introduce here, as examples of elementary parts, a few of those with the characters of which it is desirable the medical practitioner should be acquainted. Pus and blood corpuscles, cancer cells, &c., will be found in another place, and it is, therefore, unnecessary to discuss their characters here.

Epithelium.—The term epithelium (*επὶ, upon, θαλλω, to sprout*), is usually applied to those cells which lie upon the surface of membranes, as the cuticle of skin or the epithelial covering of mucous membrane, and to those which are found in the cavities of glands, continuous with the mucous or cutaneous surfaces. We may arrange epithelial cells under two heads, 1. Those that serve the part of a protective covering. 2. Those which take part in the separation or elaboration of substances entering into the composition of the secretions. The first class comprises *scaly*, *tessellated*, *columnar*, and *ciliated epithelium*, while the second includes the different varieties of glandular or secreting epithelium.

Scaly Epithelium can be readily obtained from the cavity of the mouth, and from several other situations. The nuclei constituting the bioplasm of the epithelial cells from the cavity of the mouth, are very distinct, and can always be demonstrated without difficulty, see "sputum" in chapter XIV. If the cells be placed in a solution of potash for a short time, osmose takes place, they become somewhat globular, and ultimately the cell wall dissolves. The addition of acetic acid causes the granules in the interior of the cell to become less distinct, in fact they are soluble in this reagent. The scaly epithelium from the vagina is composed of large, irregular, and often

like those occupying the tubular network of fishes."—"On the Anatomy of the Liver," 1856, page 49.

ragged cells, pl. XXIX, fig. 8. In consequence of the flattened character of the cells of scaly epithelium, portions of them will often be found folded upon each other, and creased, as it were, in various directions. The cells of the epidermis, as well as those of nail and hair, are very firm and solid masses of formed material, imbricated with one another, but they are a variety of scaly epithelium.

Tessellated or Pavement Epithelium.—This term has been applied to the cells of epithelium which form an even layer of uniform thickness, each individual cell being placed in juxtaposition with its neighbours, but not overlapping or exhibiting the imbricated arrangement often met with in the epithelial structures just referred to. The epidermis of the frog presents a beautiful example of this form of epithelium ; the inner layer of the choroidal coat of the eye, termed *the membrane of the black pigment*, the epithelium of serous membranes, of the lining membrane of the heart, arteries, and veins, and that of part of the pelvis, of the kidney, pl. XXX, fig. 1, also present more or less of this character. The nucleus, that is, the bioplasm, of the cell is usually distinct and well developed.

Glandular or Spheroidal Epithelium.—The cells are of a more or less rounded form, although in many instances, from mutual pressure, they become polyhedral. It is this form of epithelium which takes part in the process of secretion in most glandular organs. It may be readily demonstrated in the convoluted portion of the tubes of the kidney, pl. XXX, figs. 2, 10, in the sweat glands, in the secreting tubes of the stomach, in the follicles of the pancreas, in the liver, &c. The nucleus is usually well-developed, and frequently surrounded by a considerable number of minute granules, and, in many instances, small oil globules are also present.

Columnar, Prismatic, or Cylindrical Epithelium.—The general characters of this variety of epithelium may be well demonstrated by the examination of the intestinal villi, or Lieberkühn's follicles. The epithelium of the gall-bladder, of the ureters, and of the urethra, fig. 3, is of this variety. In the evacuations of cholera, the sheaths of the villi will often be found entire, and an excellent opportunity for the examination of the arrangement of this variety of epithelium is afforded, pl. XXX, figs. 7, 8.

Upon examining a cell of columnar epithelium from the intestine, it will be often observed that at its summit the cell-wall is considerably thickened, pl. XXVIII, fig. 5, pl. XXX, fig. 8. The appearance somewhat resembles that which would be produced by the presence of very fine cilia, but careful observation has proved beyond a doubt that it is not due to this cause. Kölliker has carefully investigated this subject, and thinks he has discovered very minute pores passing through the cell-wall, and apparently filled with granules of oil. But I believe the fact is otherwise accounted for. I have long been familiar with the

appearance alluded to, and have observed the thickening not only in the cells from the villi, but in other varieties of columnar epithelium. I have not been able to satisfy myself perfectly as to the existence of distinct pores. The yolk membrane (zona pellucida) of the ova of many insects, mollusks, and fishes, and probably also of mammalian animals, is perforated by a single opening, or by a vast number of minute pores, through which the spermatozoa pass, to reach the yolk within,* but these true pores have an appearance very different from that of the faint lines observed at the summit of columnar epithelium. The latter depends, I believe, upon the partly altered fatty and other materials being drawn towards the cell, layer after layer, by the action of the bioplasm, which slowly takes it up and appropriates it.

Certain forms of columnar or cylindrical epithelium take part in secretion. The bioplasm absorbs nutriment from below, and on its opposite surface undergoes change, giving off products of secretion, which accumulate in the cell, and at length escape from its orifice, pl. XXVIII, fig. 4.

Ciliated Epithelium.—There are two principal varieties of ciliated epithelium, the one consisting of small cells of nearly the same length and breadth, and the other of the prismatic or cylindrical form. Ciliated epithelium may always be obtained for demonstration from the back part of the mouth of the frog or toad, pl. XXX, fig. 5, or from the branchiæ (the beard) of an oyster or mussel. The cells must be moistened with some of the mucus taken from the same surface, or with some of the fluid in the shell surrounding the animal, or with a little clear serum. If water be added, the movement soon stops, in consequence of osmose taking place. In examining ciliary movement, it is often advantageous to suspend in the fluid the smallest quantity of lampblack or carmine, so that the direction of the current produced by the cilia can be clearly demonstrated by the movement which is communicated to the insoluble particles.

In the human subject, ciliated epithelium is found in the following situations:—On the surface of the ventricles of the brain and on the choroid plexuses; on the mucous membrane of the nose and its sinuses; on the upper and posterior part of the soft palate, and in the Eustachian tube; in the cavity of the tympanum; on the membrane lining the frontal and sphenoidal sinuses; on the inner surface of the lachrymal sac and lachrymal canal; on the mucous membrane of the larynx, trachea, and bronchial tubes; upon the os uteri; within the cavity of the uterus; throughout the whole length of the Fallopian tubes, and upon their fimbriated extremities. About seven years ago I

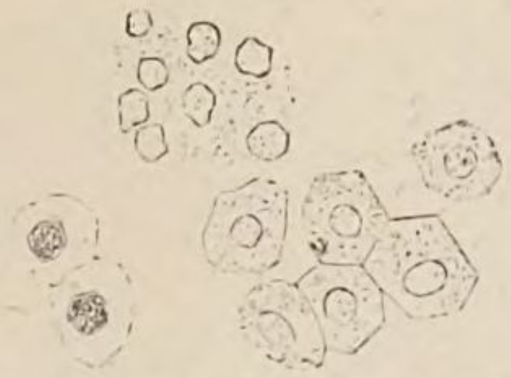
* See the very interesting observations of Dr. Ransom. Also the article "Ovum," Cyclopædia of Anatomy and Physiology, and "Micropyle," Todd and Bowman's Physiology, vol. ii, page 569.

PLATE XXX.

Fig. 1.

Tessellated epithelium from pelvis of the human kidney. p 237.

Fig 2.



Glandular epithelium from convoluted portion of the tubes. Human kidney. × 215.

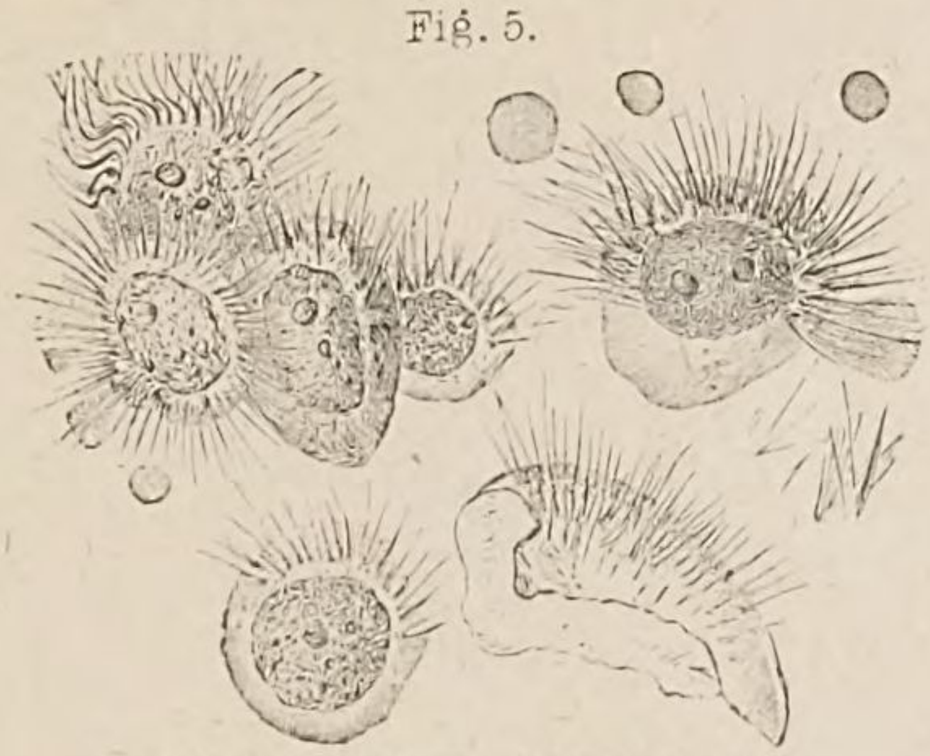
Fig. 3.

Epithelial cells of the urethra Human. p. 237.

Fig. 4.

Cells from an epithelial cancer. × 215. p. 237.

Fig. 5.



Ciliated epithelium, from the back of the tongue. Toad. × 700. p. 238.

Fig. 6.

Ciliated epithelium. Granular cells and collections of oil globules from the serum of an ovarian cyst. × 215. p. 239.

Fig. 7.

A sheath of a villus. Cholera. × 130.

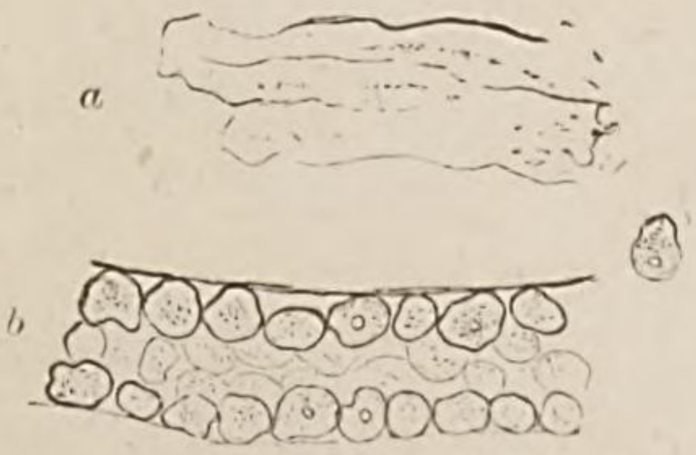
Fig. 8.

Columnar epithelium, from the jejunum of a child who died of cholera. × 700. p. 237.

Fig. 9.

Membrane. p. 213.

Fig. 10.



Basement membrane and epithelium of tubes of human kidney. pp. 237, 245.

Fig. 11.

Fibres. Fibrous appearance. p. 242.

1000th of an inch ⌊__⌋ × 215.

,, ,, ⌊_____⌋ × 700.

[To face page 238.

obtained a beautiful specimen of ciliated epithelium from the lining membrane of a large ovarian cyst, pl. XXX, fig. 6. As far as I was able to make out, the cysts in question were originally developed in the ovary, and were not formed from the Fallopian tube.

247. False Cells.—Under this head I would include all those structures which resemble, and indeed often cannot be distinguished from, true cells, but which neither "grow," multiply, nor form, nor perform any function. Many *cell-like* structures consist of granular material, oil globules, or even perfectly transparent albuminous matter aggregated together sometimes around a central mass. This might be easily mistaken for a mass of germinal matter or nucleus, by a careless observer. Indeed, the president of the British Association for 1874, following the lead of others, without having examined the grounds upon which false inferences had been based, helped further to spread the erroneous doctrine that there are living forms which differ from a fragment of albumen only by their granular character. He ought to have known that the living forms in question grow and multiply, form, and exhibit movement, while no particle of albumen or any other non-living particle whatever can do one of these things.

In many instances the central mass or nucleus of a false cell is altogether absent, and the cell seems to consist only of a collection of granules or globules. Not uncommonly, viscid matter is deposited external to an aggregation of granules or globules, and thus a sort of "cell-wall" is formed. Microscopical observers are familiar with the presence of a multitude of cell-like bodies, which consist of mere collections of oil globules, &c., and form masses varying considerably in size, and usually of an oval or spherical form. The aggregation of such particles probably depends upon physical causes, and in some instances is doubtless due to the attraction of gravitation. Strange to say, Professor Hughes Bennett, of Edinburgh, maintained long after any such hypothesis could be viewed as possible, that the living cells, pus corpuscles for example, were formed by the aggregation of granules just as false cells might be produced artificially. Professor E. H. Weber has shown that when particles of resinous substances are made to move very gradually, by the addition of a drop of alcohol to water holding the substance in suspension in a state of very minute division, and placed between two pieces of thin glass, certain currents were produced in definite directions while admixture was proceeding. At the points of rest where two such currents meet, the undissolved particles will be deposited, and thus the most regular figures, not unlike many forms of vegetable structure result.*

* E. H. Weber, "Mikroscopische Beobachtungen sehr gesetzmässiger Bewegungen, welche die Bildung von Niederschlägen harziger Körper aus Weingrist begleiten." Berichte über die Verhandlungen der k. Sachs. Gesellschaft der Wissenschaften, zu Leipzig, Math. Physisch. Classe, 1855, seite 57.

Cell-like bodies are often formed in fluids out of the body. I have very frequently observed them in solutions of organic matters undergoing evaporation. Although the solution was at first perfectly clear and free from any solid particles whatever, as evaporation proceeded, certain materials were deposited in a minute state of division. Owing probably to the motion of the fluid taking place during its concentration, these became aggregated into small collections. If I had observed these in certain fluids in my early working days, I fear they would have been set down as "cells." Some of the collections of dark granular carbonaceous matter often met with in the bronchial tubes and many forms of the so-called granular corpuscles, and similar structures met with in sputum, may be adduced as examples of false cells formed in the organism, to the characters of which, special attention should be directed. It is, however, exceedingly difficult to understand why the aggregations of oil globules, coloured particles, and other materials in a state of minute division, should attain a certain definite size and not exceed this. M. Hartig, in a paper on *aleurone*, a substance closely allied to starch, calls attention to such masses, which are seen in any liniment composed of oil and ammonia.* Such appearances are very liable to mislead, and it is the duty of every microscopical observer to study the circumstances under which such fallacies are now known to arise, and thus avoid the introduction of erroneous observations and false conclusions into science, which, having once been received as facts, especially in cases in which the course of investigation has not been described in detail, are with great difficulty afterwards corrected. Our views of "cell formation," and the growth and development of structures is now undergoing careful revision. While we have learnt to recognise the great importance of physical and chemical actions in the changes in all living beings, we are beginning to find out that the physical and chemical doctrines so eagerly embraced, and almost universally accepted in the early days of the cell theory, and even now boastfully taught as sufficient to explain all the phenomena of living beings, are utterly incompetent to account for the simplest of the phenomena which occur in the simplest living thing. And although cell-like bodies may be readily formed artificially, there is not the least analogy between these and the living cells, except in external form and general appearance. The essential part of the cell, the bioplasm or living or germinal matter, is absent from all these false-cells, and it cannot be produced artificially. This bioplasm, although perfectly transparent, we can now readily demonstrate by the use of an alkaline solution of colouring matter. See p. 65.

248. Demonstration of Cell Structures.—For the most part, cells are readily demonstrated. Care must, however, be taken that the

* "Aleurone."—"Annales des Sciences Naturelles," 1857, page 348.

medium in which they are placed does not produce a physical alteration. If, for instance, cells be immersed in a fluid, the density of which is less than that in their substance, endosmose will occur, in consequence of which the mass will increase in size, and in many instances its characters will be destroyed. On the other hand, if the density of the external medium be greater than that of the fluid in the substance of the cells, exosmose will occur, and the cell will become smaller and appear collapsed. A fluid of the specific gravity of 1015–1030, will be found to be of the proper density for immersing cells for examination; but of course no general rule can be given on this head. The fluid employed should, however, be composed of a soluble substance, which although it increases the density of the solution, has no chemical action upon the cells. Albumen, sugar, gum, and glycerine, are the most useful substances for the purpose. A good effect is often produced by a viscid solution. If glycerine, from its highly refractive properties, be undesirable, a solution of white of egg and water, or ordinary serum, may be employed. Solutions of albumen, although of very low specific gravity, are very slightly permeable. It must be borne in mind that very small quantities of syrup or glycerine have the power of increasing the density of a fluid in a very material degree, while comparatively large quantities of albumen may be held in solution without the specific gravity being much increased. Albumen, from its slight power of permeating animal membrane, is admirably adapted for the examination of delicate cell structures. A solution of albumen must be used perfectly fresh, or it may be kept from decomposition by a trace of creosote, carbolic acid, or camphor. Ordinary saliva and vitreous humour answer well as media for the examination of some masses of bioplasm, cells, and cell-like bodies.

The microscopical examination of *epithelium* does not usually present much difficulty. The surface from which the epithelium is to be taken is to be gently scraped with a knife, and a small portion then carefully removed upon the blade. If necessary, this may be moistened with a drop of water; or with a solution of sugar, or glycerine, or serum, if the cells are delicate and there is danger of rupture or endosmose. Generally, however, the addition of fluid will not be necessary. The chief reagents which will be found of use in the examination of epithelium, are acetic and nitric acids, strong and weak solutions of *potash* and *soda*, and *tincture of iodine*. Epithelium is not soluble in boiling water, alcohol, ether, ammonia, or dilute mineral acids; it is for the most part soluble in strong solutions of caustic soda and potash, and in strong acetic acid. Most forms of epithelium keep very well in the naphtha and creosote solution, in a two per cent. solution of carbolic acid, or in a dilute solution of chromic acid or acetic acid in glycerine.

Different plans for demonstrating the bioplasm of cells have been

already described, and the importance of acetic acid and alkalies in rendering the granular cell wall clear and transparent, has been alluded to. The plan of colouring *tissues* by imbibition, was first adopted by Dr. Welcker, in his researches on elastic fibres and muscles. Cells and different kinds of formed material may be stained also with colouring matter of bile, which may be easily obtained by extracting inspissated ox bile with alcohol. In cases of jaundice many cells in different parts of the body are stained of a very deep yellow colour, and cells and casts of the uriniferous tubes, where the jaundice is associated with renal disease, will be found in the urine and form very beautiful objects. The eminent advantages of a solution of carmine for colouring the colourless bioplasm only have been already referred to in p. 65.

249. Fibres and a Fibrous Appearance.—The term fibre, as applied to microscopical objects, has not been well defined. Thus, the distinct *cylindrical* elementary cords of yellow elastic tissue, have been well named "fibres," while the elementary muscular fasciculus, totally distinct from them in anatomical characters, has also been termed a fibre." This word has been applied to the delicate line-like markings seen when a band of white fibrous tissue is examined. Although the fibrous band seems to be composed of a collection of minute threads, it is impossible to separate white fibrous tissue into a number of minute individual elementary fibres. The tissue may be truly said to exhibit a *fibrous appearance* under the microscope, but it is not possible to split it into fibres of any determinate size. Most observers, however, attach a definite meaning to this word. By *fibre* is understood the elementary cords of a number of which many tissues are composed; the fibres may pass in various directions, interlace with each other, or be completely coiled up, but they must consist of the same structure throughout. In this sense, the term would seem inapplicable to the elementary muscular fasciculus. Structures presenting a fibrillar and fibrous appearance, are represented in pl. XXX. Whenever we observe lines parallel to each other, much curved, or interlacing in various directions, whatever their length may be, we speak of this as a *fibrous material*, and say that the tissue has a *fibrous texture*.

A "*fibrous appearance*" is very often fallacious—thus, a delicate membrane arranged in a number of plaits or folds, may be mistaken for fibrous tissue. Capillary vessels, when quite free from blood and stretched somewhat, have a *fibrous appearance;* but it is hardly necessary to say that no separate fibres can be demonstrated in perfectly normal capillaries.

Delicate nerve fibres in textures which are immersed in water and aqueous fluids look as if they were mere fibres. In various forms of "connective tissue" fine nerve fibres often exist in vast numbers, but are not seen if the ordinary methods of preparation are followed. The

masses of bioplasm or nuclei in connection with nerves have been often summarily disposed of as mere connective tissue corpuscles. Many of the drawings in some of the best German text books are defective in this point. Drawings in them representing nerves, capillaries, and other delicate structures, distorted by preparation, as mere "*bindegewebe.*" The matter which helps to form the basement membrane of a gland tube or follicle, and the capillary vessels and nerve fibres embedded in it, is often spoken of as "*fibrous matrix*" or as "connective tissue," but at least in many cases in which this term has been employed, the fibrous appearance has been due merely to the crumpling of the capillary walls and delicate membranous structure in consequence of stretching or pressure. If the vessels be injected with a perfectly transparent fluid, such an appearance will be no longer visible, in consequence of the thin transparent membrane of which the capillary walls are composed, being put upon the stretch. The most perfectly transparent material when thrown into longitudinal parallel folds, exhibits a striated appearance which, without very careful examination, would certainly, but most improperly, be termed *fibrous*.

In describing appearances seen in the microscope, it is important to ascertain whether the appearance produced is due to the presence of real fibres, or merely depends upon striations caused by the mode of development and growth of the tissue. This point can only be determined in many cases by very careful and patient enquiry.

250. Limitary Tissue and Basement Membrane.—Basement membrane is restricted to that clear, transparent, and excessively thin expansion, which separates the epithelium from the vessels and other structures beneath it, but which unquestionably exhibits a continuity of structure with the delicately fibrous connective tissue in which these ramify, and at least in some instances is partly constituted of nerve fibres and capillary vessels, lymphatics, and other structures. The term *limitary membrane* has also been used. The general characters and disposition of basement membrane in the different glands, was very thoroughly described by Bowman in his well-known article "*Mucous Membrane*," in the "Cyclopædia of Anatomy and Physiology," published in the year 1845.

Basement membrane is often so thin that its thickness cannot be measured. It is certainly less than the 1-20,000th of an inch. The student will find that the best organ from which to obtain a good specimen of basement membrane, is the kidney. A thin section may be cut with a sharp knife, or Valentin's knife, and after being well washed so as to remove the epithelium, the basement membrane of the tubes and the vessels will be left. Frequently empty transparent tubes may be seen projecting from the edges of the section; the membrane of which they are composed being sufficiently firm to prevent the tube from

collapsing. In the finest ducts of the liver the basement membrane is of extreme tenuity, although its presence may be satisfactorily demonstrated in injected specimens, pl. XXXI, fig. 2.

"Basement membrane" is always perfectly passive, and has certainly none of those marvellous metabolic powers some physicists are prone to confer upon it. That it results from changes occurring in bioplasm can be demonstrated if its development be carefully traced. It is *lifeless* and quite incapable of giving rise to any new structures whatever. It cannot reproduce any cells removed from its surface, but in many instances small masses of bioplasm remain upon it. It is from these that new cells are formed. The cells do not grow or sprout from the basement membrane, as used to be supposed.

Besides being spread out as a smooth expansion which is covered with epithelium, the surface of the basement membrane and structures associated with it, are frequently increased in extent by being thrown into deep projecting folds, or prolonged into little tongue-like processes which stretch from the general surface. On the other hand, there may be a folding-in of the membrane so that little crypts or follicles are formed. These are of course lined with epithelium. Such is the structure of the simplest form of *gland*. The more complex glands differ principally in the increased extent and more complicated arrangement of the interior of the follicle. Such a simple inflexion of mucous membrane increased very much in depth, becomes a *tubular gland*. When the tubes are connected together by transverse branches, we get a *network*. If the follicle be supposed to be divided into numerous small cavities by incomplete septa, while its aperture is narrowed into a constricted tube or duct, we have a *follicular gland*. If a number of these follicles be arranged together, a *conglomerate gland*, like the salivary, pancreatic, or mammary gland, is formed.

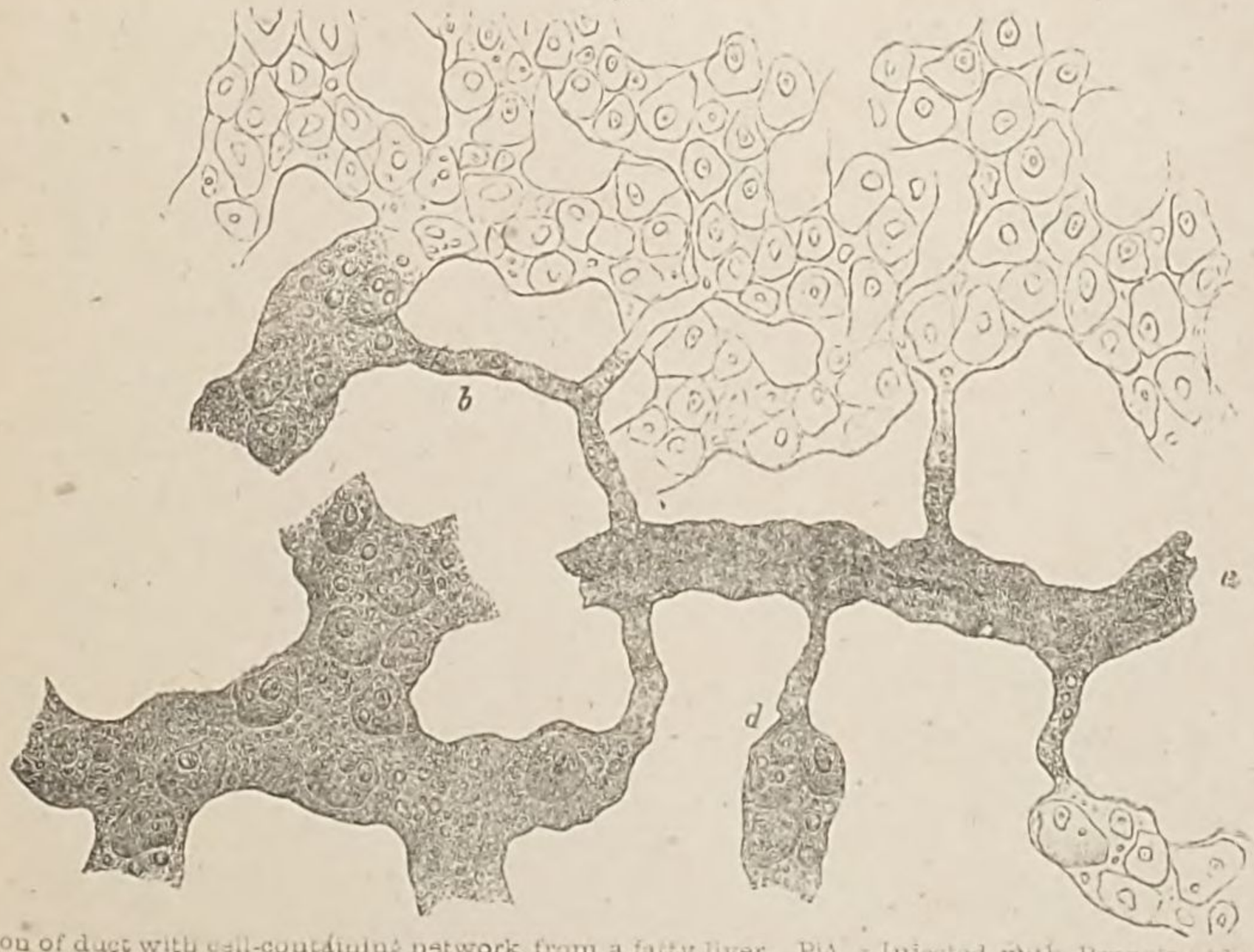
In all these cases the basement membrane takes the form of the gland. In the case of a tubular gland, like the kidney, we may remove the whole of the epithelium from the interior, and a simple tube of "basement membrane" will remain behind. This membrane intervenes between the epithelium and the capillary vessels, which are often connected with it. Through it everything separated from, or absorbed into, the blood, must pass. It has no visible holes or pores, but it is readily permeable to fluids. In many cases it permits the transudation of fluid in both directions; in some instances only in one. Sometimes it allows one fluid to pass in one direction, and another in the opposite.

There can be no doubt that basement membrane is a modified form of delicate connective substance or tissue. It is continuous with the "sub-basement" connective tissue, and when thickened, as it often is in disease, a fibrous structure can be discerned. Nerve fibres and capil-

Fig. 1.

Arrangement of muscular fibres, nerves, and tracheæ of the common maggot. The muscles represented are concerned in shortening the body. They are connected at each extremity with one of the segments. The contractile tissue of the muscular fibre in the centre of the drawing has been ruptured, and has contracted within the tube of the sarcolemma. A fine branch of the nerve trunk is seen to cross the sarcolemma and give off a still finer branch, which, after being followed for a short distance, appears lost upon the sarcolemma. If the nerve trunk be traced, many other branches distributed in a similar manner will be observed. The black lines, represented in one part of the drawing only, are the tracheæ. × 40. p. 215.

Fig. 2.



Connection of duct with cell-containing network from a fatty liver. Pig. - Injected with Prussian-blue fluid. The small trunk, *a*, gives off several smaller branches, *b*, distended by the injection which has reached the cell-containing network. × 215. p. 211.

1000th of an inch ⌴ × 40.

" " ⌴ × 215.

[To face page 244.

laries often enter into its structure and cannot be separated from it. (See my drawings of sarcolemma of muscle, pl. XXXI, fig. 1, the tubular network of the liver, fig. 2, and the uriniferous tube of the kidney represented in fig. 10, pl. XXX, and those in my memoir "New Observations on the Structure and Formation of Nerve Fibres and Nervous Centres, 1864," reprinted from the Phil. Trans.)

In disease this texture may be increased in thickness to such a degree as to become nearly impermeable to fluids which passed through it very readily in its healthy state. In a case of amyloid degeneration of the liver in which I carefully examined the thickened capillaries in different parts of the body, I was led to conclude that the thickening has been caused by the deposition of insoluble protein matter from the blood, which had been, as it were, plastered upon the inner surface of the capillary vessels. Delicate limitary or basement membrane may become granular from the deposition of albuminous, calcareous, or oily granules. Oil globules may be deposited in it or upon it. Basement membrane may be separated from the capillaries which supply it by collections of oil globules, the accumulation of fluid, or by the growth of bioplasm out of which cells are developed, which differ in every respect from those found in the same parts in health. The bioplasm may become converted into a new tissue. Basement membrane may be rendered so brittle as to give way in many places upon very slight pressure. When examined under the microscope, the specimen is seen to contain a number of pores or apertures. Such a condition occurs in the lung tissue in cases of Emphysema, as was first demonstrated by Mr. Rainey. The properties of the membrane may become so altered that a fluid which it retains in the healthy state will readily pass through it; or a fluid which passes through in the normal state with the greatest rapidity may be entirely prevented from permeating it in consequence of alteration in its texture.

251. Capillaries.—The capillaries are tubes composed of delicate membrane by which the blood is distributed to the various tissues. The material to nourish every tisssue as well as that from which every secretion and excretion is formed must pass through the capillary wall from within outwards, while the substances resulting from the disintegration of the tissues must pass in the contrary direction.

There are no actual pores in the capillary walls through which particles even very much less than blood corpuscles could make their way. But no such pores are required to account for the facts known in connection with the passage of corpuscles out of the blood-vessels. Narrow longitudinal rents or fissures are produced if the walls of capillaries are stretched. Indeed, it is possible to conceive that without any stretching whatever, particularly in cases in which the internal surface of the capillary had undergone change, a minute mass of bioplasm might slowly

insinuate itself into any slight fissures, and gradually makes its way through.

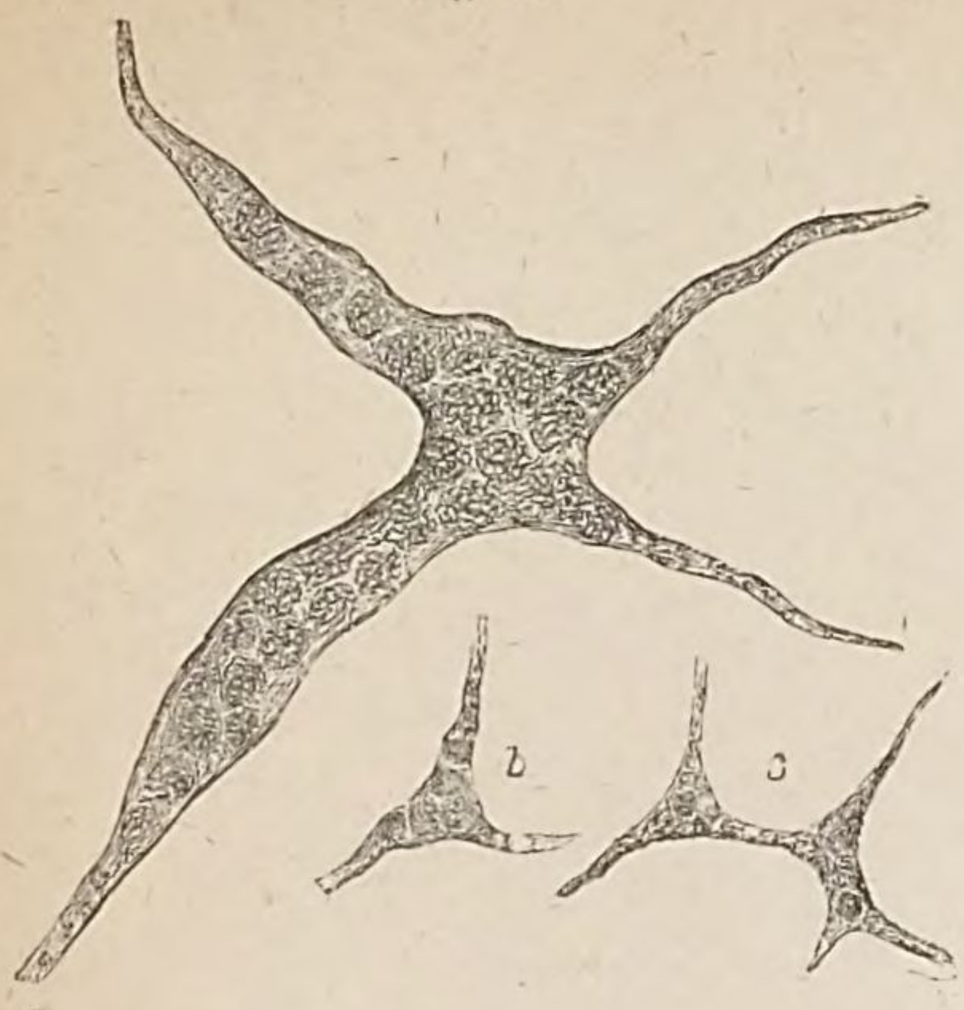
Another supposition that has been widely spread without the slightest justification is that the walls of the capillaries consist of protoplasm,* and it has been suggested that it is through this soft-yielding matter that the colourless blood corpuscles make their way towards the external surface of the vessel; where, without altering their properties, they are supposed to live on or die as pus corpuscles. There is, however, no doubt that the bioplasm of the capillary extends through the entire thickness of the vessel, and that, therefore, at these spots, and at the point where the bioplasm joins the capillary, particles would more easily pass through than in other situations.

At definite intervals, often on alternate sides of the tube, are situated the masses of bioplasm or nuclei, which are intimately concerned in the preparation of materials adapted for the nutrition of the tissues as well as in the absorption of certain substances from the tissues and their introduction in an altered form into the blood, figs. 1 to 8, pl. XXXII. I believe these masses of bioplasm may give origin to the so-called white blood corpuscles. As they alter much in size and sometimes project into the cavity of the capillary, they must affect the rapidity of the capillary circulation. When the capillary is contracted, these masses of bioplasm on opposite sides of the vessel must almost touch. Red blood-corpuscles must constantly rub against them as they are driven through the vessels, giving to or taking from the bioplasts heat and substances of various kinds. In certain cases of inflammation these bioplasts increase in size to such an extent as to interfere altogether with the passage of the blood, pl. XXXVII, figs. 5, 6. These masses of bioplasm are extremely numerous, but they vary greatly in number in different vessels. There are very many of them in the capillaries of the brain, lung, and Malpighian bodies of the kidney of mammalia, pl. XXXII, fig. 6.

The distribution of capillaries is different in every tissue, and the number of these vessels varies very greatly. Those structures in which active changes are going on, are of course largely supplied with blood, while those in which the nutritive changes are slow, contain few vessels. Cartilage and fibrous tissue are probably the least vascular tissues of the body, and their anatomical elements are separated from the blood by a considerable distance. During their active period of growth and development, however, many vessels exist. The liver is one of the most vascular organs in the body, and every part of each secreting cell is within the distance of about 1-3,000th of an inch from the blood, while the surfaces of most of the cells are only separated from it by a

* This is an illustration of the misuse of this unfortunate word. In many cases the wall of the capillary is clearly seen to be transparent membrane. We might just as well call the posterior elastic lamina of the cornea, protoplasm.

Fig. 1.



From periosteum of tooth. Young vessel with included corpuscles (white blood corpuscles). At *a* it was continuous with a small artery. *b*, a vessel at an earlier stage. *c*, two cells showing how the tube is formed. The bioplasm of the 'cell' gives rise to the included corpuscles. × 700. p. 246.

Fig. 2.

Capillary showing masses of bioplasm projecting into its interior. Areolar tissue. Mouse × 700. p. 246.

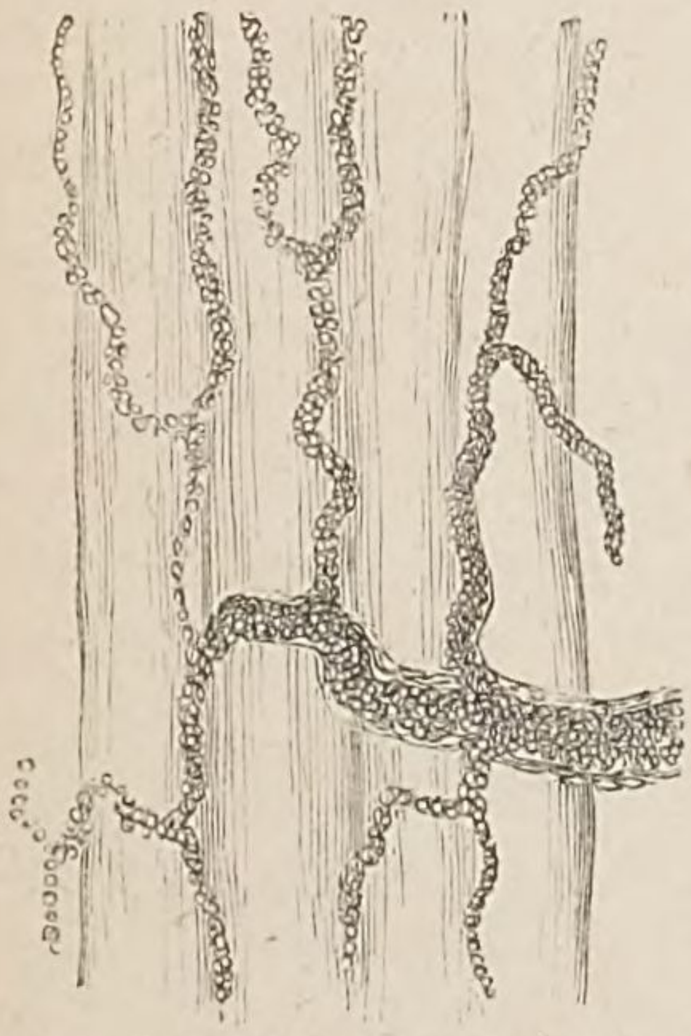
Fig. 3.

Capillary vessel from the mucous membrane of the epiglottis. Man aged 74. × 700. 172.

Fig. 4.

One of the masses of bioplasm from the interior of the capillary. Fig. 3. × 700. p. 246.

Fig. 5.

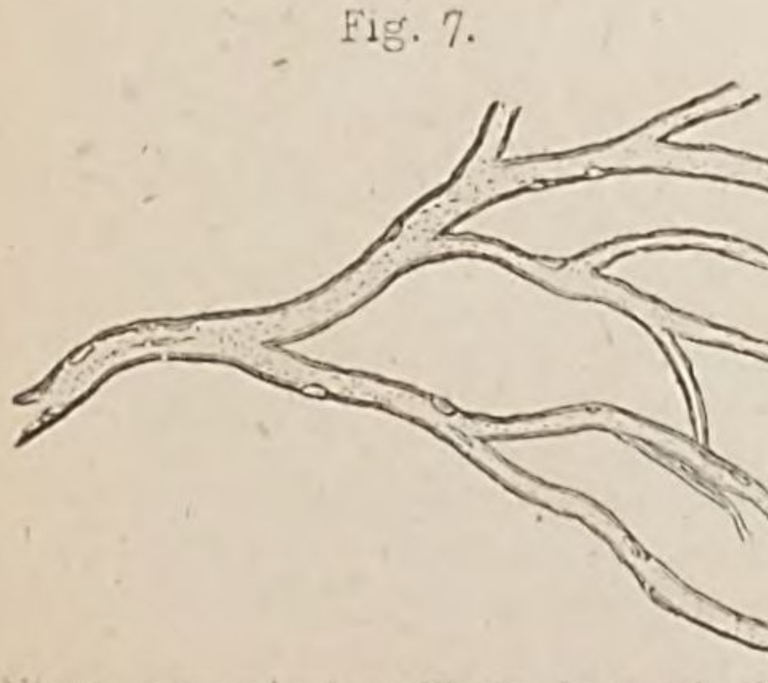


Capillaries and small vein distended with blood corpuscles. Muscle. Cattle plague. × 130.

Fig. 6.

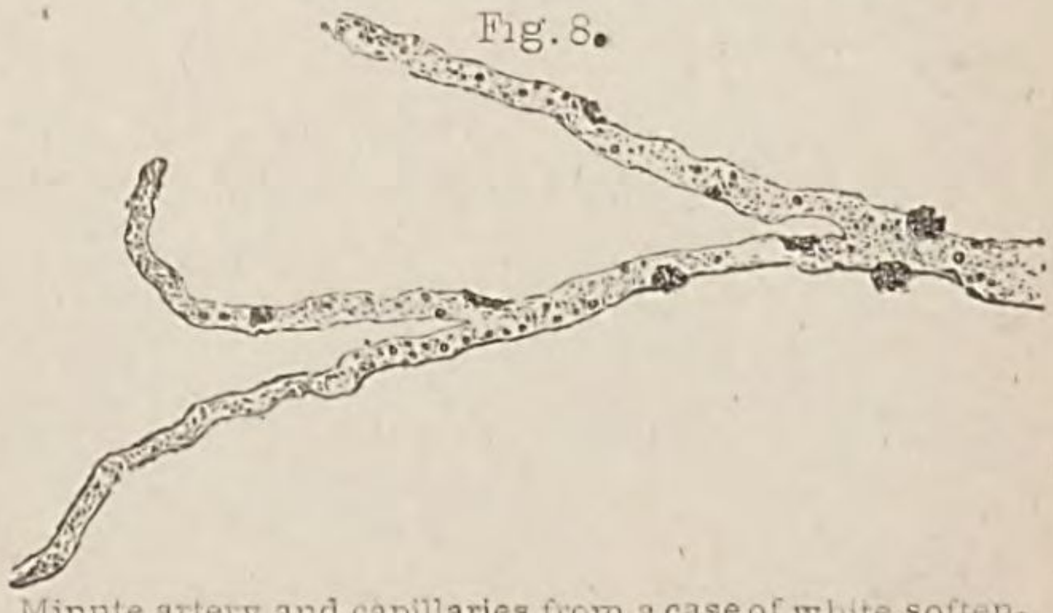
Capillary vessels from Malpighian tuft. Human kidney: showing nuclei connected with their walls. *a*, a few coils separated from the rest of the tuft. *b*, a portion of a loop showing nuclei a little flattened. *c*, tissue which connected the coils with each other. *d*, small portion of capillary pressed as much as possible to show thickness of wall at the point of reduplication. p. 246.

Fig. 7.



Minute artery and capillaries from a healthy brain. Human subject. × 215. p. 246.

Fig. 8.



Minute artery and capillaries from a case of white softening of the brain. The bioplasts of the capillary walls have degenerated, their position being shown by collections of oil globules. Human subject. × 215.

1000th of an inch ⌊ ⌋ × 130.

" " ⌊_____⌋ × 400.

" " ⌊_____⌋ × 700.

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membranous interval, certainly less than the 1-20,000th of an inch in thickness.

The capillaries cannot be properly examined unless they have been previously injected. If the observer wishes to examine their walls, or desires to ascertain the relation which they bear to adjacent parts, they must be injected with a transparent injection, and examined in fluid in as fresh a state as possible (see Chapter VI).

252. Of the Changes of the Elementary Part in Disease.—Modern research has proved most conclusively that the process of "inflammation" can affect non-vascular tissues, such as the cornea, tendon, and other fibrous textures and cartilage. This fact was demonstrated by the beautiful observations of Redfern.* In fact, the changes which take place in the vessels, important though they be in man and the higher animals, are secondary to, and often but the consequence of, changes which have been going on perhaps for some time in tissues external to them. Now, what changes can be observed in cells of epithelium during the advance of inflammation, and how are the alterations in the characters of the cells to be explained? Epithelinm may produce pus. A surface upon which epithelium exists in the normal state may very soon become converted into a pus-producing surface. It has also been proved beyond question that corpuscles, like pus-corpuscles, may sometimes be actually seen in the interior of cells. In leucorrhœa there is seldom any difficulty in finding cells of vaginal epithelium with pus-corpuscles in their interior, and in inflammation of the bladder the same fact may be observed in the case of vesical epithelium. But although these points have long been known, at least in Germany, and are generally accepted as facts, it is surprising what curious hypotheses have been resorted to in order to explain them. Buhl, of Munich, advanced the remarkable hypothesis that pus-corpuscles developed in the connective tissue corpuscles squeezed themselves, in some unexplained way, and migrated according to unknown principles, through the very narrow channel supposed to exist in the attached extremity of a cell of columnar epithelium. He supposed they might pass by the nucleus and grow and multiply in the interior of the cell! Virchow and many of his school maintain that pus can arise from connective tissue corpuscles and epithelial cells only, that in fact suppuration never occurs but in these two classes of tissues.

Pus, as I have demonstrated, consists of bioplasm or living matter, while *tissue*, that is, cell-wall, intercellular substance, &c., consists of formed material which never gives rise to living matter. In fact *Tissue*, so far from proliferating and forming pus, never proliferates at all, and is destroyed by the growth of living pus. The pus lives upon and at the expense of the tissue.

* "Abnormal Nutrition in Articular Cartilages, with Experimental Researches on the Lower Animals." 1850.

Although it has been generally admitted that pus corpuscles may be the direct descendants of epithelial cells, no one has described precisely how they are produced. Nor was it possible this could be done as long as the generally received opinion respecting the structure of cells, and the action of the cell-wall, cell-contents, and nucleus was accepted. My view differs from others essentially in this particular, that the living active part of the "cell" is defined and distinguished from the passive, lifeless cell-wall and intercellular substance. Now, in the process of inflammation it is not in fact the *cell, as a whole*, that is *actively* concerned, but only the *living part of it within*, which consists of what I have termed "bioplasm."

A normal epithelial cell takes up a certain quantity of nutrient matter within a certain time, and certain changes are produced in a definite order and in regular succession. This cell, it is said, may be excited or stimulated to *increased action*—that is, the normal process of nutrition, under certain circumstances, may be exaggerated, and the cell may take up a much larger quantity of nutrient matter within a corresponding period of time, and increase in size with greater rapidity than before. To say that the exaggerated action is the result of the "irritation" is surely no explanation at all. We find as a fact that the cells grow faster, and take up more nutrient matter, soon after a foreign body has been forced into close contact with them, so as to displace or injure them; but, not only are the cells that are touched influenced, but all those in the immediate neighbourhood.

The drawings in pl. XXXIII will enable the reader to understand what occurs. In all the drawings, the roundish granular mass represents the *living matter* or *bioplasm*, and the outer faintly shaded layer the *formed material*, which was once bioplasm. Now, in the normal state, nutrient pabulum gradually passes through the formed material into the bioplasm. Certain constituents of the pabulum immediately acquire the same wonderful powers as the bioplasm already existing, while the particles of bioplasm matter upon the surface of the mass undergo change and are resolved into the formed material of the cell-wall, and other matters which pass away. This formed material is always formed *from within*, as explained in p. 231, so that the layers first produced are pushed outwards by the formation of new matter within. This may be incorporated with that which was first produced, or several successive layers may be formed one within the other. The bioplasm is the only *formative, living, growing, or active part of the cell.*

Suppose, then, such a cell as represented in fig. 1, *a*, pl. XXXIII, to be supplied with an increased quantity of nutrient pabulum, what changes will be observed? The outer hardened formed material may be torn or ruptured mechanically, as in a scratch or prick by insects (fig. 1, *c*), or it may be rendered soft and more permeable to nutrient pabulum by

the action of certain fluids. In either case it is clear that *the access of pabulum to the bioplasm is facilitated*, and the latter necessarily "*grows*,"—that is, converts certain of the constituents of the pabulum that come into contact with it, into matter like itself. The mass of bioplasm increases in size, as in fig. 1, *b*, and soon begins to divide into smaller portions. Parts seem to move away from the general mass, fig. 2, *d*. These at length become detached, and thus several separate masses of bioplasm, which are embedded in the softened and altered formed material, result, fig. 2, *e*. If the formed material was ruptured, the bioplasm would of course soon escape, leaving the remains of the cell-wall (formed material of the original cell) behind. The masses of bioplasm increase in size, and even live at the expense of the softened formed material, which was formed from the original mass of bioplasm, and at length escape, fig. 2, *f*, *g*. The *free masses* of bioplasm now in contact with the pabulum grow and multiply rapidly. Each forming upon its surface a more or less viscid material, which corresponds to the formed material of the cell, but is softer and much more readily permeated by nutrient matter.

In this way the so-called "inflammatory product," *pus*, results. The *abnormal pus-corpuscle* is invariably produced from the *bioplasm or living matter of a normal epithelial cell, in consequence of the bioplasm of this cell being supplied with pabulum much more freely than in the normal state.* The change as it occurs in epithelium, is seen in fig. 4, pl. XXXIII.

I have arrived at these conclusions from studying the changes which actually occur in specimens coloured with carmine, by which the bioplasm can in every case be readily distinguished from the *formed material.* The nature of the changes occurring in cells in inflammation can then be explained easily enough if the artificial nomenclature of *cell-wall*, *cell-contents*, *nucleus*, be given up. In all acute internal inflammations a much larger quantity of inanimate pabulum is taken up by certain cells and converted into living matter than in the normal state. Hence there is *swelling*, *increase in bulk.* Cells of particular organs, which live very slowly in health, live very fast in certain forms of disease. More pabulum reaches them, and they grow more rapidly in consequence.

Next, as to the changes which occur in cells which *have been growing very rapidly* and are returning to their normal condition, in *which the access of nutrient pabulum is more restricted than in the abnormal state.* These changes nearly correspond with the ordinary changes which occur in many normal cells as they pass from the embryonic to the fully formed state. The outer part of what was a free mass of bioplasm undergoes conversion into formed material, and this increases as the supply of pabulum becomes reduced within the ordinary limits.

Instead of pabulum being rapidly taken up by the mass of bioplasm,

as was the case when it was growing rapidly in the abnormal condition, and the division of the mass into several smaller portions within a very short time, the amount of pabulum taken up bears a nearer relation to the proportion of formed material produced, and as this latter slowly accumulates, and the entire cell becomes larger, the absorption of nutrient matter becomes less and less; partly because the cell is farther removed from the nutrient surface, but mainly in consequence of the progressive increase in thickness of the formed material which separates the living bioplasm from the nutrient fluid which surrounds it. These changes are represented in figs. 1, 2, pl. XXVII, p. 232.

Next, let me consider the case of soft cells, or elementary parts, like the "liver cell," the "*formed material*" of which under *normal conditions* becomes *quickly* resolved into other soluble constituents. If such cells be placed under circumstances which cause the formed material to become harder and less permeable to nutrient matter than in health, what alterations will be observed in the character of the cell?

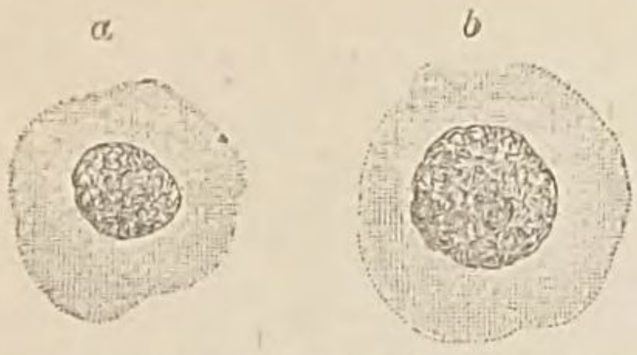
In health the whole of the material of which the liver cell is composed, is soft, moist, and readily permeable to certain nutrient matters. There is no *cell-wall*, but as has been already remarked, the outer part of the formed material is gradually resolved into soluble biliary matters. which pass down the ducts, and saccharine matter which permeates the walls of the vessels and enters the blood, pl. XXVIII, fig. 6, p. 232. To make up for the disintegration of the outer part of the formed material, new formed material is produced in the interior of the cell from the bioplasm, which as it undergoes change is replaced by new bioplasm produced from the pabulum that is absorbed.

Now, if such cells and their descendants are bathed with improper pabulum, and especially with substances which render albuminous matters insoluble, or possess the property of hardening them, they necessarily diminish in size, and the work performed by them becomes less. In other words, in consequence of the formed material becoming less permeable, less nutrient matter is taken up, and of course as the formed material becomes hardened, less disintegration takes place, and the quantity of secretion which really contains the products resulting from the disintegration of the formed matter, is much diminished. Under the supposed conditions the cells shrink much in size and become much harder. Many gradually waste, and not a few completely disappear. These are the changes which take place in the liver cells during the progress of cirrhosis, and to these changes in the cells the shrinking, and condensation of the whole organ, so characteristic of this disease, are due. Fig. 3, *a*, pl. XXXIII, may represent a normal liver cell, and *b* and *c* will give some idea of the cells in different stages of condensation and shrinking. *See* also pls. XXVII, XXVIII, p. 232.

From the foregoing observations it follows that disease may result in

PLATE XXXIII.

Fig. 1.



Increase in size of bioplasm in an adult epithelial cell, from increased access of pabulum. p. 248.

Rupture of formed material, permitting free access of pabulum. pp. 232, 248.

Fig. 2.

FORMATION OF PUS. To illustrate the changes in the bioplasm of an epithelial cell resulting from increased nutrition. Showing the manner in which the bioplasm of a normal cell, if supplied very freely with pabulum, may give rise to pus. p. 249.

Fig. 3.

Condensation and hardening of soft formed material, with shrinking of the entire cell, as occurs in the liver cells in cirrhosis. p. 250.

Fig. 4.

A portion of the epithelial covering of the tongue in a state of inflammation. The bioplasm of the cells at the lower part of the drawing is growing and multiplying very rapidly. The changes occurring in individual cells are shown to the right of the figure. × 619. p. 249.

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two ways:—either from the bioplasts of an organ growing and multiplying faster than in the normal state, or more slowly. In the one case, *the normal restrictions under which growth takes place are diminished; in the other, the restrictions are greatly increased.* *Pneumonia* is a good example of the first condition, for in this disease millions of bioplasts are very rapidly produced in the air cells, and nutrient constituents are diverted from other parts of the body to this focus of morbid activity. *Cirrhosis* is a good example of the second condition, for an organ in which great and rapid changes occur normally, and in which an enormous quantity of nutrient matter is constantly taken up and converted into new material, gradually becomes quiescent; the amount of change becomes less and less, the whole organ wastes, and the secreting structure shrinks; and at last inactive connective tissue alone marks the seat where most active and energetic vital and chemical changes once occurred. It is easy to conceive how such a substance as alcohol must tend to restrict the rapid multiplication of the bioplasts in the first condition, and how it tends to promote the advance of such a condition as cirrhosis.

Inflammation consists essentially in the very rapid growth and multiplication of masses of bioplasm. The general appearance and some of the properties of bioplasm are the same in all cases, and we could not distinguish the bioplasm of a low plant or animal from that of the human brain cell. In man and the higher animals it is the slow growing formed material alone which gives to the anatomical elements of tissues their individual peculiar characters. When growth is so rapid that there is not time for the production of this formed material, there must of necessity be a great difference in appearance between the rapidly multiplying masses of bioplasm occurring in inflammation (pus corpuscles), and the normal cell, the bioplasm of which gives origin to them. In the lowest and simplest organisms there is no state exactly resembling what we call "inflammation" in the higher, because in them the freer access of an increased supply of pabulum is followed merely by increased multiplication of the ordinary very simple cell or elementary part without any departure from its normal characters. *This fact very clearly indicates the true nature of the inflammatory process.* In some very simple organisms, however, a difference *is* to be observed between the structures which are produced *very quickly* and those which are *more slowly* formed, for in these, just as in higher forms of existence, *time* is required for the full development of the perfect tissue. *Inflammation* does not occur in any of the lower forms of microscopic fungi; but when pabulum is abundant, minute and simple organisms are alone produced, while when the supply becomes more restricted this rapid multiplication ceases, and a more perfect structure grows from the lower than from the more rapidly increasing forms. In fact, by altering the

conditions under which even such simple organisms as these grow, we can, indeed, cause *rapid multiplication of the masses of bioplasm*, which will be surrounded by a very thin layer of formed material, or by restricting pabulum, we may *promote the production of a vast amount of "formed material."* (See a paper on the Growth of Common Mildew in Vol. II of my Archives, p. 179, 1861.)

CHAPTER XIII.

LYMPH, CHYLE, BLOOD, SEROUS FLUIDS.—*Examination of Blood.—New Researches.—Size of the Red Blood Corpuscles.—On Estimating the number of Red Blood Corpuscles.—Of the white or colourless Living Blood Corpuscles or Blood Bioplasts.—Of Microcytes and of Lostorfer's Corpuscles.—Blood in Disease.—Blood in Lower Animals.—Of Examining Blood Stains in Medico-Legal Investigations.—Detecting the Blood Corpuscles.—Hæmin Crystals.—Guaiacum Test.*—SERUM.—*Examination of Serous Fluids.—Fluid from Serous Cavities.—Fluid from Cysts.—Fluid of Ovarian Disease.*

253. Lymph and Chyle.—A drop of lymph or chyle may be subjected to examination in a thin glass cell. Chyle can he obtained very readily from the thoracic duct or from the lacteals of an animal which has been fed with fatty matter for two or three hours before death (page 135). The character of the corpuscles should be observed; first, with a quarter of an inch object glass, and then with high powers, and the reaction of the corpuscles with acetic acid studied.

Besides the well-known lymph and chyle corpuscles, which are nearly as large as white blood corpuscles (page 258), there are masses of bioplasm in lymph and in chyle, which are so extremely minute and transparent that they will certainly be passed over if the examination is conducted with ordinary powers, including even the *one twelfth of an inch* object glass. Such particles are, however, of the highest importance, and well worthy of the most attentive examination. They consist of small portions of bioplasm or living matter which once formed part of a chyle or lymph corpuscle, but which have been detached, and are in fact growing larger, and will, at length, form new corpuscles. It is remarkable that when separation has once taken place the particles never again coalesce, but as long as the new corpuscle continues attached to the parent by the narrowest pedicle, it may be again drawn into the mass. In order to observe these points it is necessary to employ a $\frac{1}{25}$ or a $\frac{1}{50}$ of an inch object glass. By the use of these very high powers not only do we gain the advantage to be derived from increasing the apparent size of a body, but we may be able to see that which before

was not visible. Objects which from their extreme tenuity are quite invisible to ordinary powers, magnifying as much as 700 diameters, may be seen very distinctly with the aid of these highest objectives. And extremely delicate masses of bioplasm, not to be detected by ordinary examination, are highly important. They are, indeed, the active agents concerned in the absorption of nutrient matters from without, and by their increase and multiplication only can new lymph and chyle and blood be formed. The small quantity of fibrin present in these fluids is formed by the bioplasm only, pl. XXXVI, figs. 6, 7, 8, 9. Perfectly pure lymph may sometimes be obtained for examination from a cutaneous lymphatic trunk, which opens upon the surface of a wound or ulcer. Such cases, however, are not commonly met with.

Cases have been recorded by Dr. Carter, Dr. Buchanan, and others, in which the condition of chylous urine was associated with a dilated state of the lymphatics of the leg and thigh, actual chyle being discharged from these lymphatic vessels. *See* "Kidney Diseases, &c., 3rd edition, p. 305. In pl. XXXIV, figs. 9, 10, the chyle-like fluid from a remarkable case of Dr. Day's, is represented.

254. Examination of Blood.—In order to examine the blood, a small drop is placed upon a glass slide, and covered with thin glass, which is to be pressed down until a very thin, transparent, and almost colourless stratum only remains, care being taken not to completely crush the corpuscles. If in this manner the individual globules cannot be distinctly seen, a little serum, or white of egg and water must be added; but it is better to avoid the addition of any fluid, if possible. Upon carefully focussing, the *red globules* will appear to present a dark centre and light circumference, or the reverse, according as the focus is altered, pl. XXXIV, figs. 1, 2, 3, pl. XXXV, and pl. XXXVI, fig. 7, and here and there a white corpuscle may be observed.

The *white corpuscles* are rather larger than the red, and the largest and oldest have a granular appearance. Upon the addition of acetic acid, from one to three nucleus-like bodies (new centres of growth) make their appearance in the white corpuscles, and not unfrequently these may be seen without the addition of any reagents.

If a little strong syrup be added to a drop of blood, the red corpuscles will become much flatter from exosmose of a part of their fluid contents; while, on the other hand, if placed in water, they become spherical from endosmose, and may swell up to such an extent as to be perfectly transparent and invisible. It is not difficult to make a saline solution of similar density to that of the red blood corpuscle, in which it will not alter in form at all. In this manner, as Dr. Rees expresses it, we may "take the specific gravity of a blood corpuscle."

Acetic acid causes the red corpuscle to become very transparent and clear, and to swell up. After the application of this reagent, the

blood corpuscle may be scarcely visible, but the material of which it is composed is not dissolved. Cold strong hydrochloric and nitric acids do not immediately dissolve the globules; by the latter reagent the outline is often rendered darker and thicker, while the entire globule is caused to shrink. Red and white blood corpuscles are entirely soluble in ammonia and in alkalies. They are rendered darker, and the walls corrugated, by the acid of the gastric juice; and, after remaining in acid urine for some time, a similar change occurs; hence the black colour of blood, which has been effused into the stomach, and the dark smoky hue of acid urine containing blood. This smoky hue is especially distinct in cases in which the blood has escaped from the uriniferous tubes, and has thus been gradually but very intimately mixed with the urine. Blood crystals, and the method of obtaining them, have been described in § 230, p. 191.

255.—New Researches on the Blood Corpuscles (1864).—The facts referred to so far can be demonstrated by the magnifying powers in ordinary use, from 100 to 300 diameters, but it is not too much to say that in consequence of recent investigations with much higher powers, our views concerning the nature of the blood corpuscles and the changes taking place in the blood have been completely altered. An almost entirely new field for elaborate and highly important physiological and pathological enquiry has been recently laid open. I propose to advert very briefly to some facts which seem to me of great importance, and which, if followed up, will certainly lead to the discovery of new lines of highly important investigation.

Of the Red Blood Corpuscles.—The red blood corpuscle of man and mammalia generally consists of a mass of soft viscid matter, perhaps of the consistence of thick treacle, composed of hæmato-crystallin. It is, at least in certain states, soluble in water, but is only dissolved by serum and the fluid part of the blood very slowly. The outer part of this matter is of firmer consistence than the interior, especially in the older corpuscles, but there is no special envelope or cell-wall. When the latter are placed in water the more soluble matter is dissolved, leaving the harder external portion. By the action of many chemical reagents the outer part of the red blood corpuscle is condensed. The appearances thus caused, and others, have led observers generally to the conclusion that the red blood corpuscle was a "cell," in fact, an envelope or cell-wall enclosing fluid contents of a different composition. So firm has been the conviction that this was so, that the rupture of the "cell," and the escape of the contents have been spoken of as if this had actually occurred, and the external membrane of the ruptured corpuscle has been described as if it had been seen. That the red blood corpuscle is not a cell is proved conclusively by the following facts:—

1. A red blood corpuscle may be divided into many smaller portions,

every one of which assumes the spherical or spheroidal form, and in many cases become stellate, pl. XXXIV, fig. 3.

2. The mass of bioplasm in the case of the "nucleated" blood corpuscle of the frog and some other vertebrates, or a portion of it, may pass right through the red viscid material of which the outer part of the corpuscle is composed without the rupture of any membrane, just as a solid particle might pass through treacle or molten pitch, pl. XXXIV, fig. 5, *a*, *b*, *c*.

3. A red blood corpuscle from Guinea pig's blood assumes the crystalline form very readily, and without the addition of any reagent. The process may be watched under the microscope, and a single corpuscle may be clearly seen to become a single crystal, or by the application of a gentle heat a corpuscle may be broken up into several smaller portions, every one of which becomes a tetrahedral crystal, pl. XXXIV, figs. 6, 7, 8.

4. Several red blood corpuscles under certain circumstances run together, forming a soft homogeneous viscid mass, in which nothing like cell-walls or the remains of such structures can be seen, and which undergoes crystallization in every part without exhibiting indications of cell-walls anywhere, fig. 6.

5. When water is added to blood corpuscles, they swell up just as a piece of jelly would do, but they do not burst as is generally stated. No doubt soluble matters are dissolved out, but as the water evaporates, the corpuscles assume their previous form, although they appear paler than before. Although many appearances may be urged in favour of the existence of a cell-wall, the above facts and those learnt from studying the changes taking place during the development of the corpuscle, seem to be absolutely incompatible with such a view. Neither here nor elsewhere is the 'cell-wall' a necessary or essential portion of the elementary part. A thing may arise from a thing which existed before it, grow, live, perform its offices, and increase its kind, without having a cell-wall at any period of its existence. Cell-walls may be easily made artificially. In spite of the importance hitherto attached to it, the 'cell wall' is of no special significance.

It is generally stated that the red blood corpuscles of an animal exhibit a certain definite size, but it will be found that they vary extremely, so that corpuscles exist of various dimensions. If we examine blood with the highest powers, not only do we meet with extremely minute corpuscles, but many of these are so very transparent that they could not have been seen at all under a lower power. Extremely transparent bodies are demonstrated under high powers, which would certainly be passed over by those in ordinary use. The reader is referred to pl. XXXVI, fig. 7, in which the various corpuscles met with in healthy blood are represented. It is probable that the very small pale corpuscles

PLATE XXXIV.

Fig. 1.

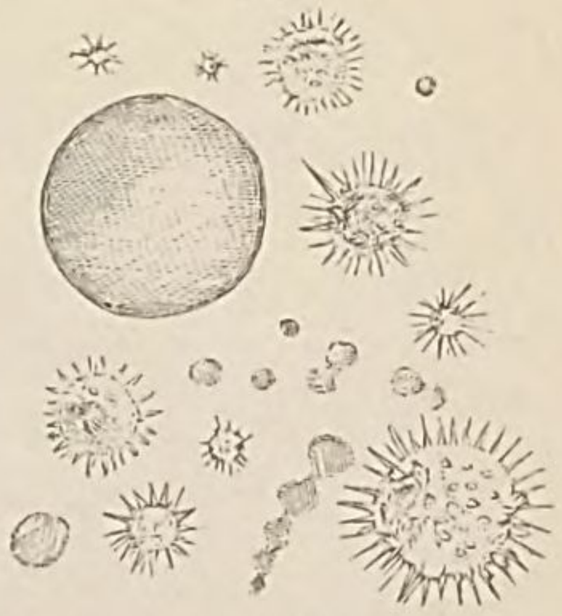
Blood corpuscles from healthy human blood. × 215. p 254.

Fig. 2.

× 215

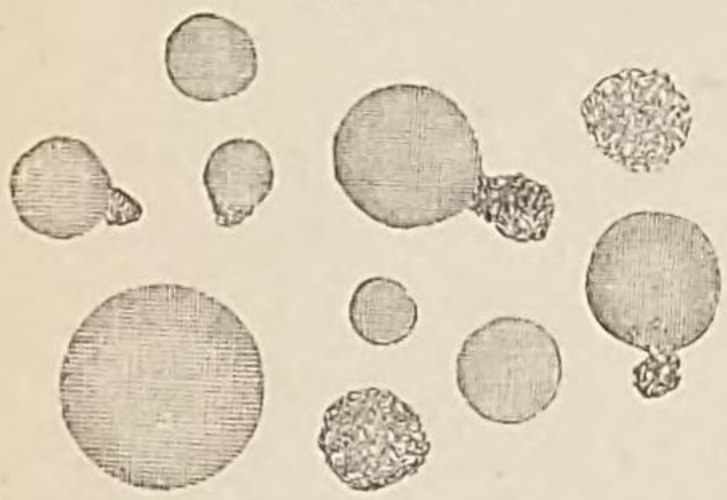
Human blood corpuscles *a*, *b*, *c*, from the living body; *d*, *e*, *f*, from the urine. × 215. p. 257.

Fig. 3.



Human red blood corpuscles, dividing into spherical and stellate particles after pressure. × 1800. p. 254.

Fig. 4.



Colourless bioplasm, separating from formed material of young human blood corpuscles. × 1800. p. 258.

Fig. 5.

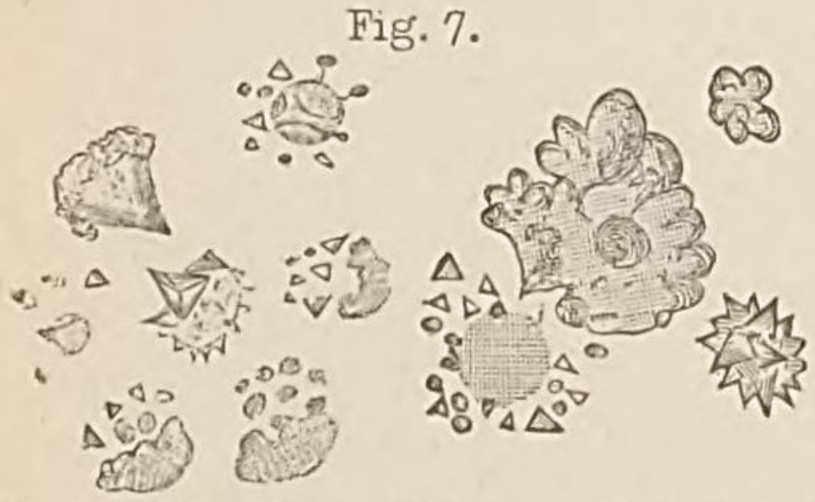
Portion of living bioplasm tending to separate into smaller portions.

Portions of living bioplasm which have divided into smaller portions, some of which have passed through the formed material into the surrounding fluid. p. 256.

Fig. 6.

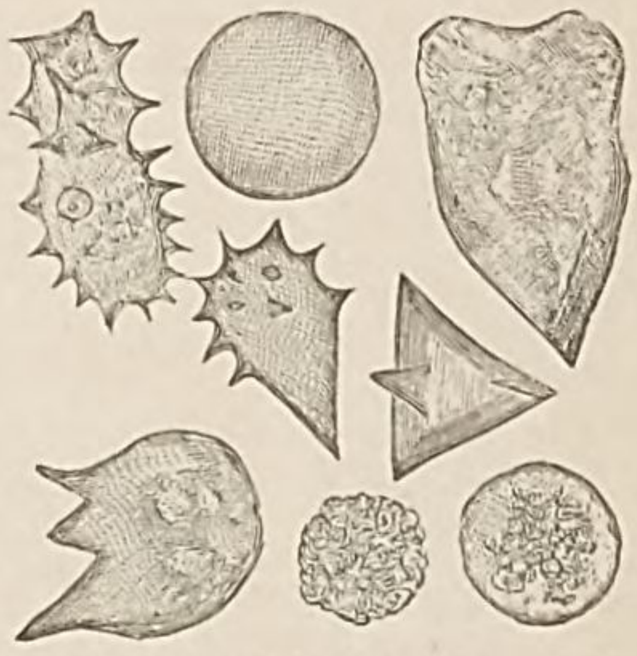
Blood crystals from the blood of the guinea-pig. × 700. p 256.

Fig. 7.



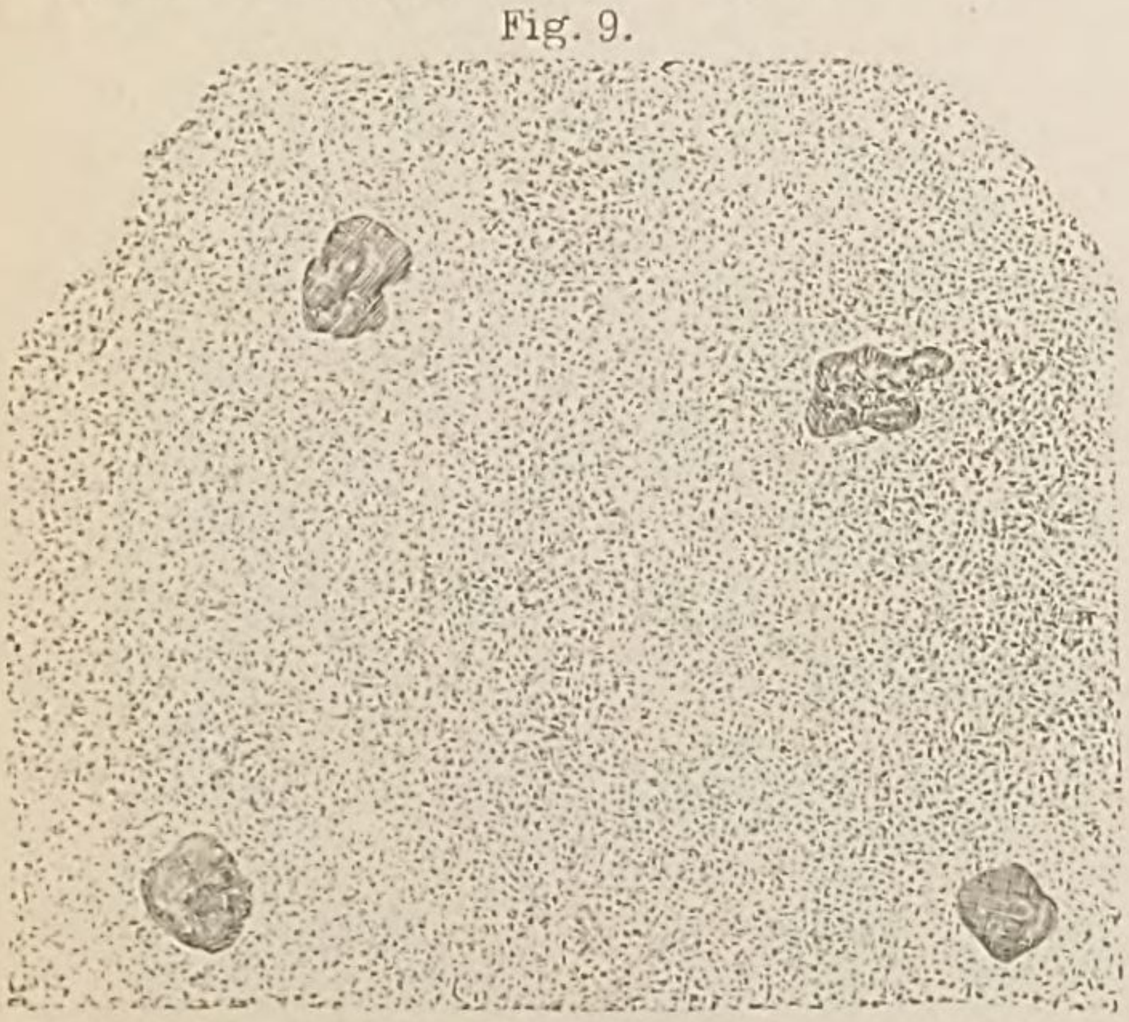
Disintegration of red blood corpuscles of guinea-pig's blood, and formation of crystals, after application of a gentle heat. × 700. p. 256.

Fig. 8.



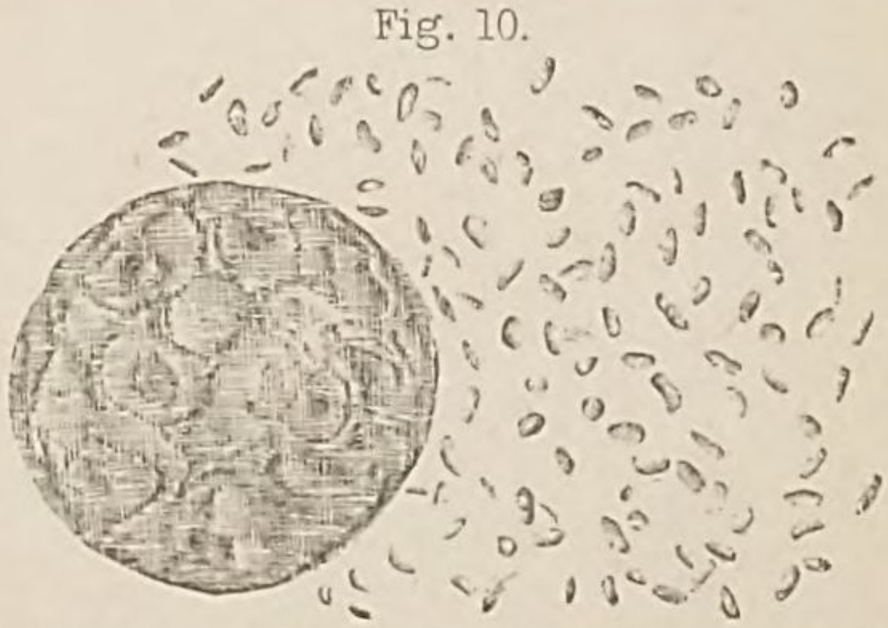
Formation of blood crystals from the red blood corpuscles of guinea-pig's blood, shortly after removal from the body. No reagent added or heat applied. × 1800. p. 256.

Fig. 9.



Chyle. From an enlarged lymphatic of the prepuce of a child. × 700. 1869. p. 253.

Fig. 10.



Chyle. Same as Fig. 9, but magnified by the $\frac{1}{5}$ × 2000. p. 253.

1000th of an inch |—| × 215.

" " |———| × 700.

" " |———————| × 1800.

[To face page 256.

seen in this figure are young blood corpuscles which are gradually undergoing change, and acquiring colour and the characters of the red corpuscles. The colourless bioplasm of living matter of which they are at first composed, gradually becomes converted into the coloured formed material, pl. XXXVI, fig. 1, *a*, *b*, *c*.

Red blood corpuscles often assume a stellate form, pl. XXXVI, fig. 7, pl. XXXVII, fig. 2, which is not very easily explained. As this has been observed in certain cases of disease it has been regarded by some as a morbid change. Not only is it commonly observed in the case of corpuscles found in perfectly healthy blood, but these may be divided and subdivided into very small portions, every one of which will exhibit the stellate appearance, pl. XXXIV, fig. 3. Nor do I think that this change can be due to alterations occurring in bioplasm, for although masses of bioplasm undoubtedly do often assume a stellate form, the red blood corpuscles of the Guinea-pig become stellate soon after their removal from the vessels, and certainly in some cases the sharp spine-like projections formed become the angles of tetrahedral crystals. See pl. XXXIV, fig. 8.

256. Size of the Red Blood Corpuscles.—The blood corpuscles of hundreds of animals have been accurately measured by Mr. Gulliver, who has shown that different orders or families of mammalia are characterised by different grades of corpuscles. The largest blood disks among mammalia are found in the edentata. Those of the orycteropus, myrmecophaga, and elephant are nearly of the same size, notwithstanding the great difference in size of the animals. In the family of rodents the largest occur in the capybara, and the smallest in the tiny harvest mouse. If now a rodent existed as large as an elephant, his blood corpuscles would be of enormous size. Mr. Gulliver's memoirs are published in the Proceedings of the Zoological Society, June 11, 1844, February 10, 1870, and in other numbers.

The valuable figures printed in plate XXXV were kindly sent to me by Mr. Gulliver, who corrected some of his former drawings and added some to those which he originally published in 1862, in a paper which was presented to the Zoological Society. See "Proceedings of the Zoological Society," February 25, 1862. The specimens from which the drawings were taken were in most instances obtained from the living animal, a small quantity of the blood being allowed to dry upon a glass slide. All the figures are magnified equally, or nearly so. By a glance at this valuable plate the observer will at once realise the extraordinary variations in dimensions which characterise the blood corpuscles of certain vertebrates closely allied to one another, and the equally remarkable uniformity in size which obtains in certain cases between creatures far removed.

257. On Estimating the Number of Blood Corpuscles.—This opera-

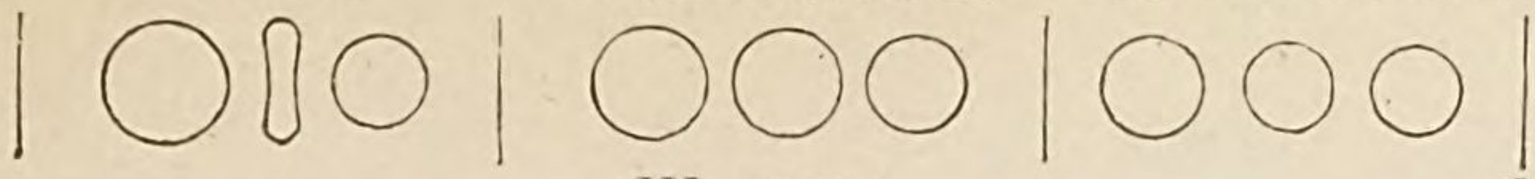
tion may be effected roughly by placing a drop of blood upon a glass slide, and pressing very firmly upon it a small piece of thin glass so as to obtain the thinnest possible stratum for examination. Upon examining this with a quarter, an approximative idea of the number of corpuscles in a small area which has been carefully marked out, may be formed. If specimens of the blood of patients suffering from various diseases be examined in this way, the greatest differences in the number of the corpuscles will be observed. Vierordt has proposed a plan for determining the number of corpuscles in a given quantity of blood numerically, by the microscope, and Welcker has improved upon this. It is obvious in such very delicate researches the slightest error will become very considerable, when, from the data obtained by the above method, the number of corpuscles in a large quantity of blood is calculated. The operation is a very delicate one, and requires great care. As a full description of it would occupy much space, I think it better to refer those who desire to employ it to the original memoirs, than to give a short summary of the plan which would be useless to anyone who desired to practically employ it.*

258. Of the White or Colourless or Living Blood Corpuscles.—The general nature of the white blood corpuscles has been already referred to, and the student has been recommended to study the vital movements which take place in them during life. It has been shown how protrusions occur which become detached, and thus from one living corpuscle several minute particles of bioplasm matter may be derived. Many of these, which are less than the $\frac{1}{10000}$th of an inch in diameter, may be seen in blood if a $\frac{1}{50}$th object glass be employed. These minute particles of bioplasm may ultimately assume the characters of the ordinary full sized colourless corpuscles,—or passing into the current of the circulation, and being exposed to the influence of the respiratory and other processes may continue to grow, and at the same time undergo change into red blood corpuscles. The living bioplasm becomes gradually resolved into the red lifeless hæmato-crystallin, which accumulates, and probably into other substances, which escape. The hæmato-crystallin is probably diffused through the bioplasm, and this latter being perfectly transparent and colourless, cannot in the mammalian corpuscle be distinguished. If, however, young red blood corpuscles be allowed to die, it will be found that shortly before death a remarkable change occurs, the diffused bioplasm separates and moves away, leaving the lifeless coloured material behind, pl. XXXIV, fig. 4. As the red corpuscle advances in age, more and more bioplasm becomes resolved into formed material, until at last the red corpuscle consists of

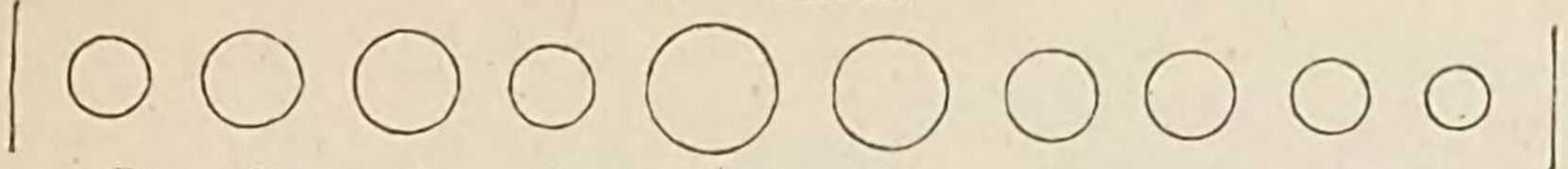
* Vierordt in "Vierordt's Archiv," Jahrg. II, Heft I. Dr. Welcker in "Archiv. des Vereins für gemeinschaftliche Arbeiten zur Förderung der Wissenschaftlichen Heilkunde," vol. i, page 161.

APYRENEMATA.

I. MAN. II. QUADRUMANA. III. CHEIROPTERA.

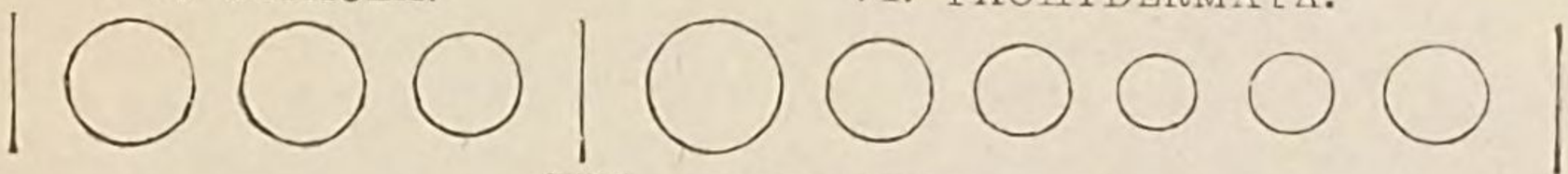


IV. FERÆ.

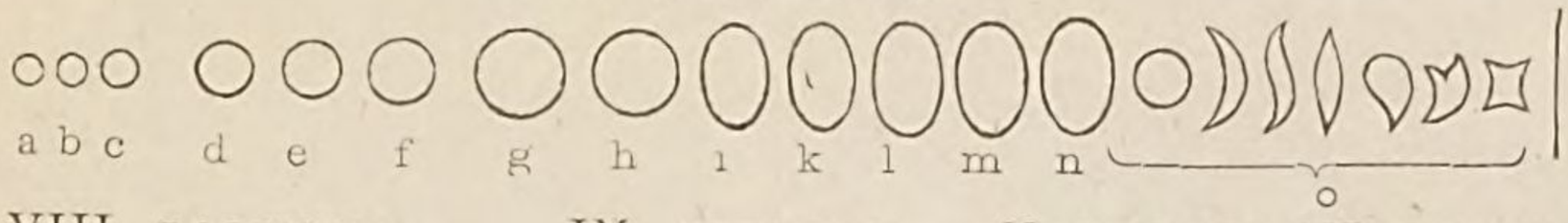


p q r s t u w x y z

V. CETACEA. VI. PACHYDERMATA.

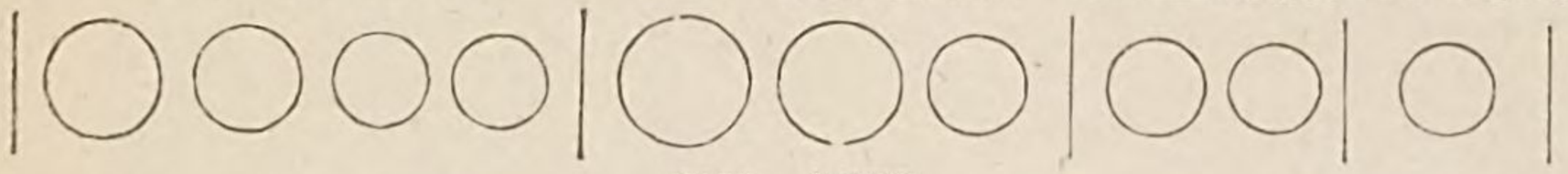


VII. RUMINANTIA.

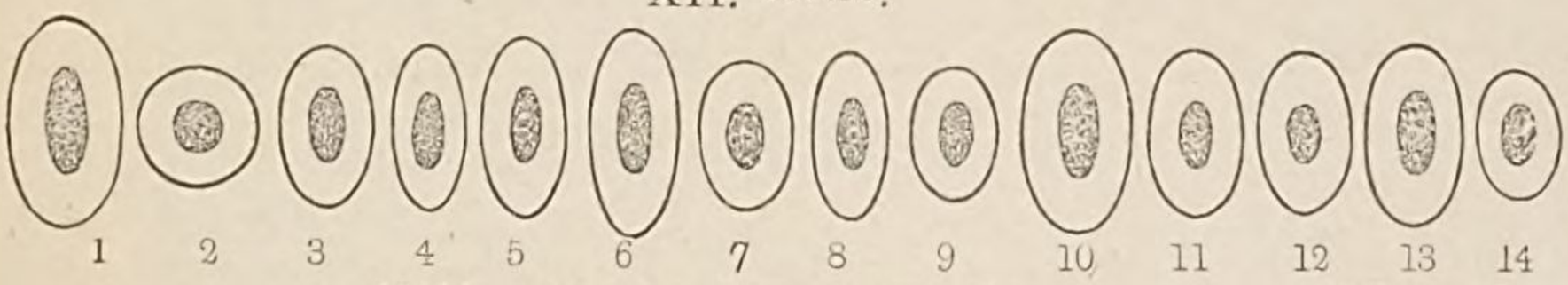


a b c d e f g h i k l m n o

VIII. RODENTIA. IX. EDENTATA. X. MARSUP. XI. MONOTR.



XII. AVES.

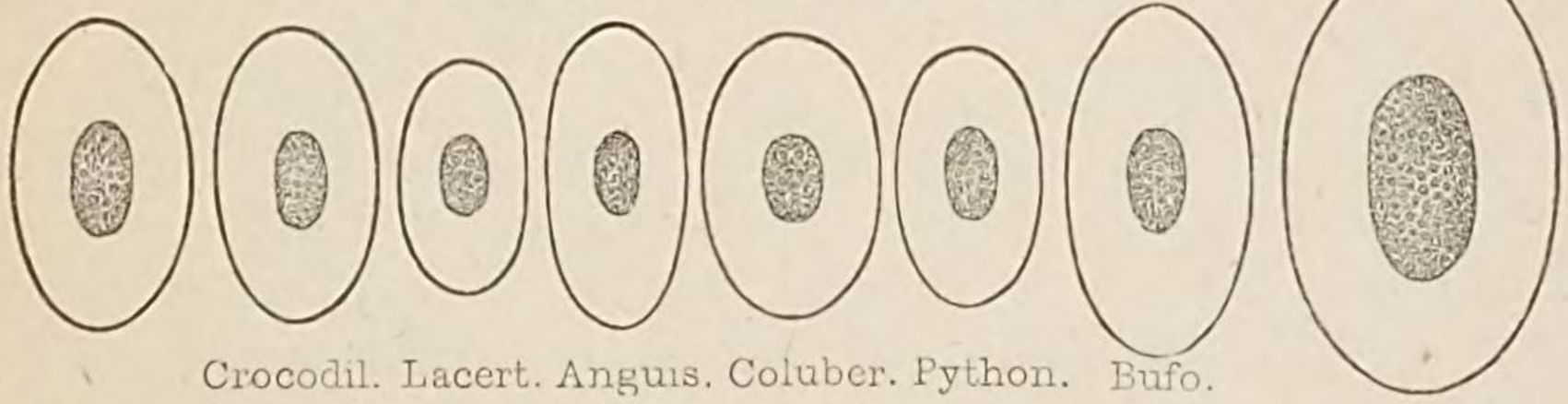


1 2 3 4 5 6 7 8 9 10 11 12 13 14

VERTEBRATA.

XIII. REPTILIA ET BATRACHIA.

Gymnopodus.



Lissotriton.

Crocodil. Lacert. Anguis. Coluber. Python. Bufo.

PYRENÆMATA.

XIV.

Sieboldia.

XV.

Siren.

XVIII.

XVI.

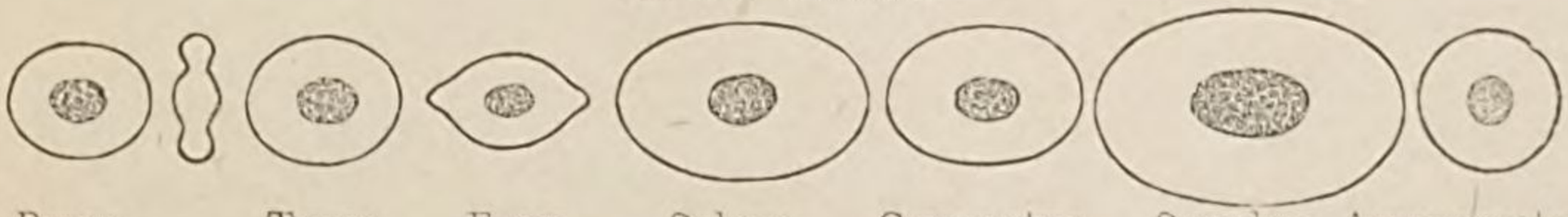
Proteus.

XVII.

Amphiuma.

Lepidosiren.

XIX. PISCES.



Perca. Tinca. Esox. Salmo. Gymnotus. Squalus. Ammocœtes.

$\frac{1}{1000}$th of an inch × 900

$\frac{1}{10000}$

G. Gulliver ad. nat. del. 1839-74

[To face page 258.

the latter substance only. The whole is then entirely dead and is slowly subjected to physical and chemical changes, gradually disintegrated and at last dissolved, and converted into new compounds.

In the nucleated red blood corpuscles of the frog, the bioplasm remains as the nucleus, but it is easy enough to find corpuscles which contain a mere speck of living matter only, pl. XXXVI, fig. 2, while others are almost entirely composed of it, being merely invested as it were with a very thin coating of the coloured matter, pl. XXXVI, fig. 1, *a*, *b*, *c*, *d*.

White blood corpuscles are no doubt derived from the lymph and chyle corpuscles by the formation of offsets or protrusions, and I believe that the masses of bioplasm which project into the interior of the capillary vessels also give origin to them, pl. XXXII, figs. 3, 4, p. 246. I cannot too strongly recommend the advanced student to study these matters, for many of the points referred to are of the highest importance in connection with many diseases, especially pyæmia and the whole class of contagious fevers. It seems to me probable that living particles of contagium gaining entrance into the blood, affect the growth or modify the changes occurring not only in the young blood corpuscles, but in the bioplasm of many tissues, and thus the numerous secondary phenomena which are familiar to us may to some extent be explained. So also it seems possible that the influence these morbid particles exert upon one generation of young bioplasts may affect succeeding ones for a longer or shorter period of time. In this way the immunity of the individual to subsequent attacks of the same disease is perhaps to be accounted for.

I have considered these points which are here only alluded to most cursorily, in two papers, "On the Nature of the Red Blood Corpuscle," and "on the Germinal Matter of the Blood," read before the Microscopical Society, December 9th, 1863. See also "Disease Germs," second edition, 1872.

The whole subject of the blood corpuscle is fully discussed in a memoir of upwards of 160 pages, by Professor Arthur Boettcher, in vol. xxxvi, of Virchow's Archiv, p. 342. In this paper a full historical account of observations on the red blood corpuscle will be found, and the researches of various observers, including those published in the first of my papers, are ably and carefully criticised.

259. Of the so-called Microcytes and of Lostorfer's Corpuscles.— Bodies have been described in certain specimens of blood which have been called "microcytes." These are less than blood disks, highly refractive, with a sharp outline. Dr. Sidney Coupland (*Lancet*, August 3rd, 1872) describes some corpuscles, of a red colour, $\frac{1}{4500}$th of an inch in diameter, in the blood of a case of Addison's disease. They disappeared as the patient improved in health.

Bodies like fungus sporules are not unfrequently found in the blood of the lower animals. Altered red blood corpuscles sometimes very closely resemble the sporules of fungi. Bacteria and bacteria germs are to be detected in the blood in various forms of low fever, and increase enormously shortly before the patient's death. They have, however, nothing to do with the causation of disease.

Sarcinæ have been stated to exist in blood. See a paper by Dr. Ferrier, "British Medical Journal," January 27th, 1872.

Great interest was excited in the year 1870, by some observations by Dr. Lostorfer of Vienna, who ascertained that he could distinguish the blood of syphilitic persons from that of health by the presence of certain peculiar bright bodies, which were developed in the syphilitic blood, in from one to five days after it had been taken from the patient. I express no opinion on the nature of these bodies, but consider it desirable to insert the following translation of one of Dr. Lostorfer's memoirs, from the "Medizinische Jahrbücher," 1 Heft, 1871, which has been copied from the half-yearly abstract of the "Medical Sciences:"—

"The great mistrust with which, in almost all quarters, observations on certain appearances in the blood in infectious diseases are received, induces me to commence this time with the results of my work hitherto obtained, and a statement of the manner in which I have shown that I can distinguish the blood of a syphilitic from the blood of a non-syphilitic individual.

"Professor Stricker, who was kind enough to put my assertion to the proof, gave to me on several occasions a number of prepared blood tests, concerning the origin of which I was ignorant, some of these having been taken from healthy and others from syphilitic persons. They had been numbered and noted in writing. After three, four, and at the most six days, I made him acquainted with the results of my examination; and then a comparison with the notes showed that—except in those instances where one or more preparations, either immediately after they were made or in the course of the examination, had been found useless, and so placed on one side—there was accordance between these notes and my results. Those tests which had been taken from healthy men I indicated as the blood of non-syphilitics, and those from syphilitic patients as the blood of such individuals.

"In a similar manner I have successfully carried out with success a test put by Professor Hebra.

"The manner in which I have carried out my investigation is as follows:—A drop of blood obtained by a puncture in the skin is transferred as rapidly as possible to a smooth slide, and covered by a piece of thin glass. The blood test thus obtained is now placed in a bell-glass, arranged as a moist chamber, in which there is a stand for twelve

Fig. 1.

Young red blood corpuscle not yet coloured. Frog. × 1800. p. 257.

b

Young red corpuscle. Frog. Formation of coloured portion. × 1800

c

Young red corpuscle. Part of coloured portion fully formed. × 1800.

d

Young red corpuscle, coloured portion and nucleus. × 700. pp. 259, 265

Fig. 2.

Old red corpuscle. Frog. Nearly the whole of the bioplasm has been converted into colourless formed material. p. 259.

Fig. 3.

Division of very young corpuscles (white corpuscles) and formation of outer coloured portion.

Fig. 4.

'Nucleus,' 'nucleolus,' and outer red formed material. p. 265.

Fig. 5.

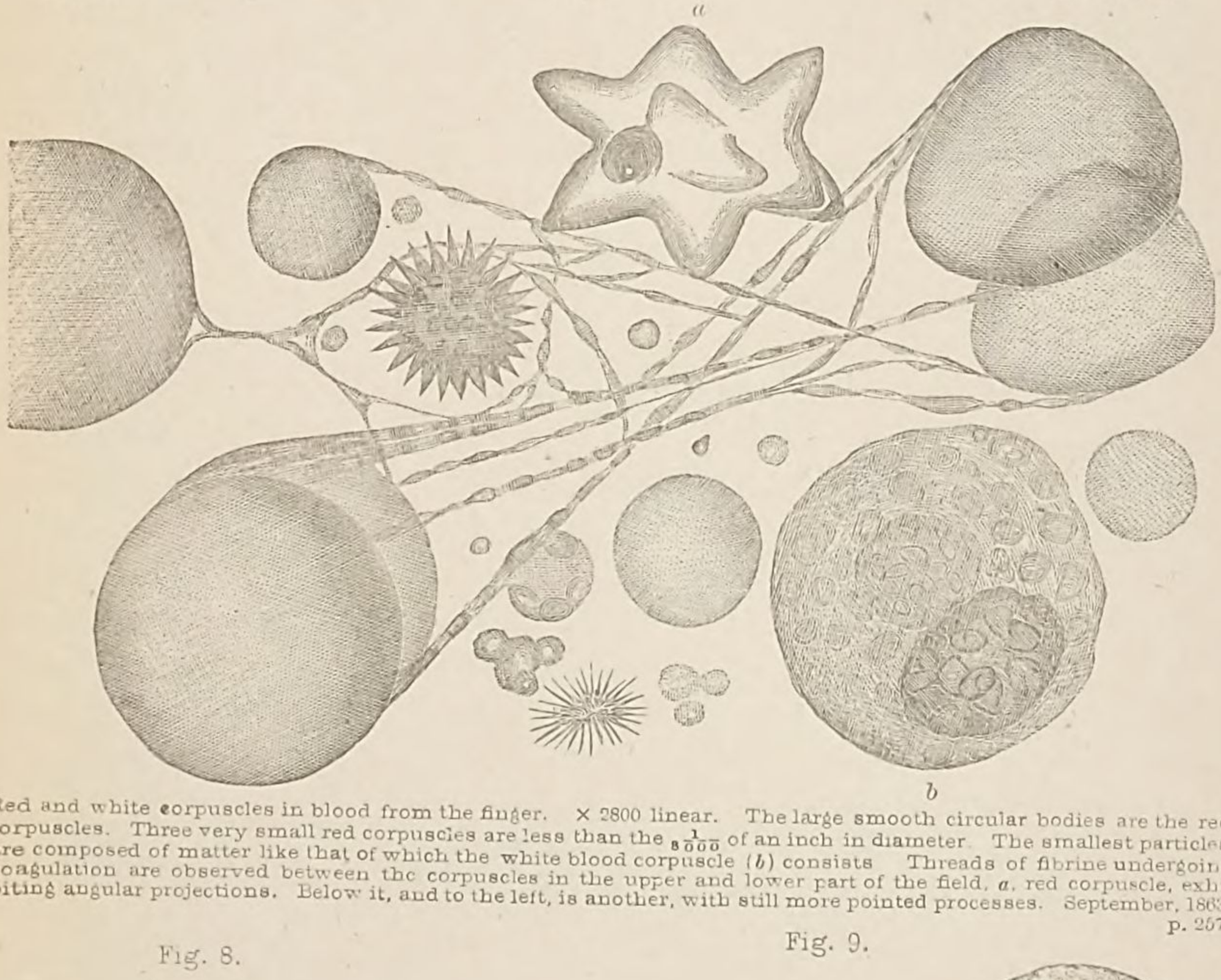
Movement of bioplasm towards surface of corpuscle.

Division of bioplasm or nucleus of corpuscle.

Fig. 6.

Portion of a large mass of fibrine from guinea-pig's blood the instant coagulation had occurred. × 1800.

Fig. 7.

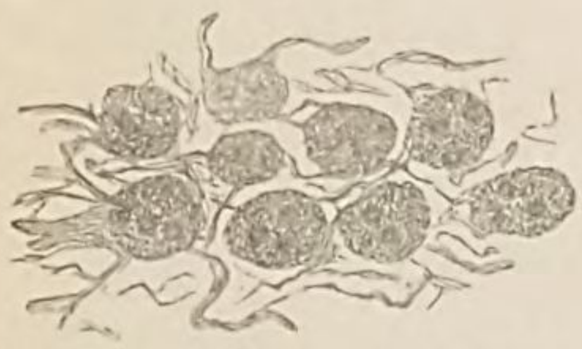


a

b

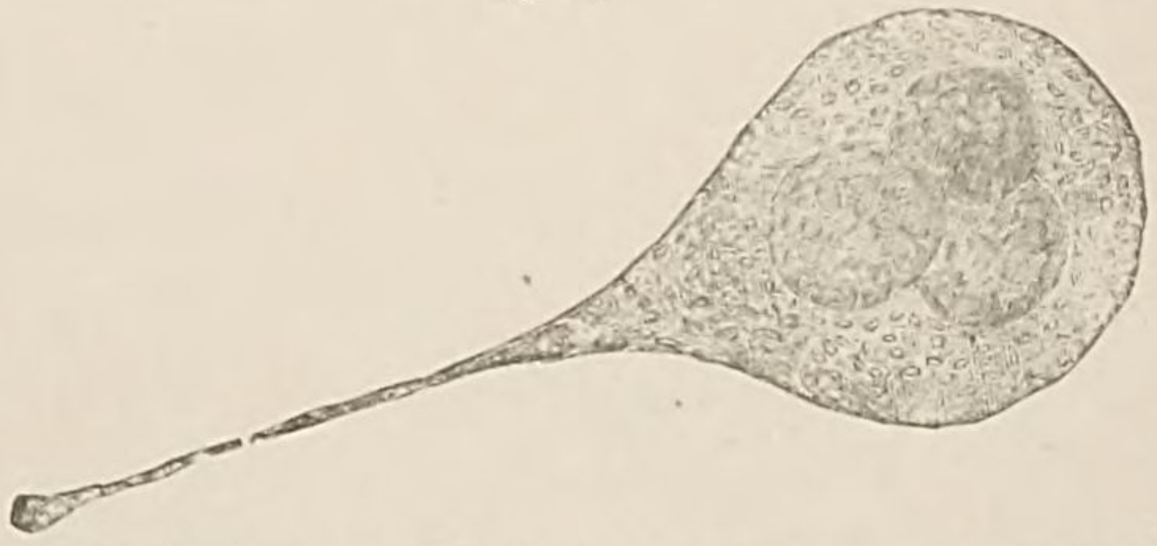
Red and white corpuscles in blood from the finger. × 2800 linear. The large smooth circular bodies are the red corpuscles. Three very small red corpuscles are less than the $\frac{1}{8000}$ of an inch in diameter. The smallest particles are composed of matter like that of which the white blood corpuscle (*b*) consists. Threads of fibrine undergoing coagulation are observed between the corpuscles in the upper and lower part of the field, *a*, red corpuscle, exhibiting angular projections. Below it, and to the left, is another, with still more pointed processes. September, 1863. p. 257.

Fig. 8.



From a pale clot in the heart of a patient who died of exhaustion, showing white corpuscles and fibres of fibrine. × 700.

Fig. 9.



White blood corpuscle (human subject) with a thread of fibrine being formed from it. × 1800.

$\frac{1}{1000}$ of an inch ——— × 700.

" " ——— × 1800.

$\frac{1}{10000}$

" " ——— × 2800.

[To face page 260

preparations. In every glass of this kind I place the blood tests both of syphilitic and non-syphilitic individuals.

"The examination of these preparations is made daily with a N. 10 oc. 3, Hartnack's immersion lens.

"Whilst, in general, during the first two days, nothing of a foreign nature was to be seen beyond some vibriones, bacteria, and sometimes the early forms of sarcina, on the third, many times on the fourth or fifth day, and exceptionally after the first twenty-four hours, I found small bright bodies, some of which were at rest and others presented vibritile movements. On some of these bodies a small process could be made out.

"On the fourth, exceptionally on the third, fifth, or sixth day, these bodies had become larger and increased in number. Many of the enlarged bodies presented the above-described process, which proved clearly to be due to a process of budding. In some, this had increased so much in size that it was almost as large as the maternal body.

"On the following days, the bodies increased more and more in size, so that some soon reached or even exceeded the size of shrivelled red blood corpuscles. At the same time, however, smaller corpuscles were present in all gradations of size. Bud-formation also was now a frequently observed phenomenon, and many corpuscles had not only one outgrowth but several, which, some with and others without a stalk, were fixed to the maternal corpuscle. There were many instances in which one outgrowth was the bearer of secondary buds. Not all of these corpuscles were circular; many were irregular in form. After the specimen of blood had been kept for eight or ten days, a vacuole was formed in each of the larger corpuscles, which enlarged more and more, so that at last it was surrounded by only a thin membrane, indicated by a double contour. At this stage the development of the corpuscles had reached its end, and no further changes were observed, even in those cases—which, however, were very rare—where the blood remained suitable for examination at the end of a month.

"I proved also the behaviour of the corpuscles on the addition of various fluids—such as solutions of sugar, distilled water, Pasteur's fluid, a solution of common salt, and a solution of acetic acid. These fluids, on the first day of their addition, had, notwithstanding their diversity, the same action. The corpuscles became shrivelled, and ran together into a quite irregularly-formed and opaque mass, and all further development was arrested. This change occurred with the greatest rapidity on addition of solution of sugar and of Pasteur's solution.

"At a later period, say about six or eight days after the appearance of these corpuscles, shrivelling occurred on the addition of one of the above-mentioned fluids, but by no means to such an extent as before. Many of the corpuscles appeared to be folded and had acquired an

irregular form. Others retained their circular form, but were smaller, their contours were more distinct and their brightness not so well marked. I have observed, however, a further development on the addition of distilled water, of an extremely diluted solution of sugar, and, in one case, of a solution of salt. In those instances the vacuoles enlarged rapidly, budding occurred, and there was also a formation of long extended processes which resembled very much the germ sacs of fungi.

"With regard to the quantity of the corpuscles in question; this varied very much: in some instances they were much scattered, in others they were very numerous. In one instance I counted on the fourth day, fifty in the field of the microscope. Whether the number of corpuscles has any relation to the existing syphilitic symptoms remains to be determined by further investigation.

"The deviations from the above described modes of development consist chiefly in a more rapid or a slower development. In the first instance, I found on the fourth day corpuscles which had attained almost the size of shrivelled blood corpuscles. In cases of this kind, the corpuscles undergo the most manifold changes of form. The bud-formation was extremely unimportant. In later cases the corpuscles became distinctly perceptible for the first time on the fifth day; their growth was very slow, and when fully attained was rapidly followed by dissolution.

"Although I had first examined the blood of many healthy men without finding any of the above described corpuscles, I still persevered in the examination, and during a period of three months, I made use of, simultaneously with specimens of blood from syphilitic subjects, specimens from healthy, or at least non-syphilitic persons, and recently blood from patients affected with gonorrhæa, ulcers, and eczema. Generally the blood of syphilitic was kept in the same chamber with that of non-syphilitic persons. I have recently examined blood specimens of several patients suffering from typhus and lupus, and also one with Elephantiasis Græcorum.

"Now, since in no blood specimen from a non-syphilitic person could I, notwithstanding the most careful examination, find any of the above described corpuscles, therefore must the possibility of finding these, after several days' incubation in the blood of syphilitic persons, be regarded as somewhat characteristic of this disease. From this time I would give to those corpuscles the provisional name of *Syphilitic Corpuscles.*

"I have not yet carried on investigations at all times of the year, but have had occasion to remark that special attention ought to be paid to the temperature. Many of my researches were made at an indoor temperature fluctuating between 10° and 18° R., others in a place where

the fluctuations of the temperature were still more considerable. In low temperatures I found that the results were negative."

In thirteen cases in which the blood of the syphilitic patients contained the corpuscles, a full account is given.

260. Blood in Disease.—The best way of examining blood is to place a drop on a glass slide, cover it with thin glass, and at once submit it to microscopical examination, but where this is not possible, the blood may be collected in the fine capillary tubes now used for vaccine lymph. These tubes may be easily made by drawing out a piece of glass tube in the flame of a spirit lamp. The capillary tube may be broken into pieces about three inches in length. The drop of blood at once runs up the tube by capillary attraction. A space must, however, be left unfilled. Each end of the tube is then to be hermetically sealed, care being taken not to permit the heat to boil the blood, otherwise it will be driven from the tube by the sudden expansion of the vapour set free. To prevent this accident, the tube should not be filled to within $\frac{3}{4}$ of an inch of each end.

In looking at a drop of healthy blood, besides the red corpuscles, here and there a larger white or colourless corpuscle is seen. The relative number of these should be carefully noted, as in disease they are liable to increase enormously. In health there is one white corpuscle to about fifty red ones. The condition in which they are much increased in number is frequently associated with enlargement of the spleen (not lymphatic and mesenteric glands), and has been termed "Leukhemia;" or, more correctly, "Leucocythemia," or "white cell blood disease," by Professor Bennett. In extreme cases, white or colourless corpuscles are almost as numerous as the red, and they appear much more so, because the red blood corpuscles collect together in little piles, while the white remain separate and distinct, and occupy the intervals or spaces thus formed. The surrounding fluid sometimes contains granules, and vast numbers of extremely minute corpuscles may be discerned by the highest powers. Upon being treated with acetic acid, the colourless corpuscles swell up a little, become more transparent, and usually display, one, two, or even more roundish bodies in the centre, much resembling those developed in the pus globule, by the action of the same reagent.

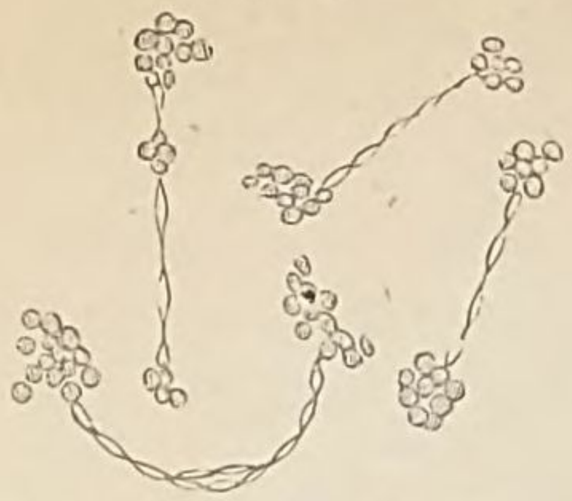
In some cases of cholera, I have found in the blood several cells, much larger than the white corpuscle. It is probable that these remarkable bodies are closely related to the white or colourless corpuscles. In one case which I had an opportunity of examining, many of these large cells contained oil globules, collected together in one part, pl. XXXVII, fig. 3. I have also seen very large colourless blood corpuscles in some cases of pyæmia. Many of these large masses of bioplasm, looking like enormous white blood corpuscles, could not certainly traverse the smaller capillary vessels.

Sometimes blood corpuscles adhere together with unusual tenacity. Of this I met with a very unusual example in the year 1854. The case was that of a man aged twenty-six, who was suffering from kidney disease. The corpuscles in the defibrinated blood manifested so great a tendency to cohere, that they collected in small masses, floating in a clear serum, and looked like minute dots to the eye, when a thin stratum of the blood was examined. By pressure they were made to separate, but soon adhered again. In this case, there certainly appeared to exist an *attraction* of the corpuscles for one another. It seems impossible to explain the facts by supposing an alteration in the density of the serum and corpuscles, pl. XXXVII, fig. 1. The adhesion of the blood corpuscles above referred to is met with in many cases of cholera, but I think the following observation renders it probable that it has no special relation to that disease, but is due simply to the blood being deprived of much of its water. One day I took some Epsom salts which produced three very copious liquid stools. I examined a drop of blood from the finger, and found the corpuscles adhering exactly as represented in fig. 1. I then took three tumblers of warm water, and in less than an hour after the first observation was made, the blood corpuscles exhibited their ordinary characters forming the piles of disks, but no longer exhibited the remarkable tendency to adhesion.

But the most interesting, and probably by far the most important of the changes yet observed in blood in disease, is the presence of a number of masses of bioplasm and products resulting from their death and decay, which are not present in the healthy blood, pl. XXXVII, fig. 2, *b*. There is reason for thinking that the particles which gave origin to these obtained entrance from without and made their way through the thin capillary walls, and thus became mixed with the circulating fluid. By their multiplication in the capillaries circumscribed local congestions are caused, and in this way peculiar 'eruptions' and 'rashes' result, figs. 4, 5. In many cases the congestion ends in complete stagnation, followed by suppuration (boil, carbuncle, pustule), and the death, destruction, and removal of the portion of tissue affected; or it is followed by the escape from the blood and lymphatics of serum and small particles of bioplasm which multiply for a time in the substance of the cuticle, the superficial portion of which is elevated (vesicle, bulla), the fluid and corpuscles drying up and forming with the altered cuticle and secretion of the sebaceous glands, a *scab* or *crust*,—or a raw moist surface, which does not readily heal, known as an *ulcer*, is formed beneath the detached layer of cuticle.

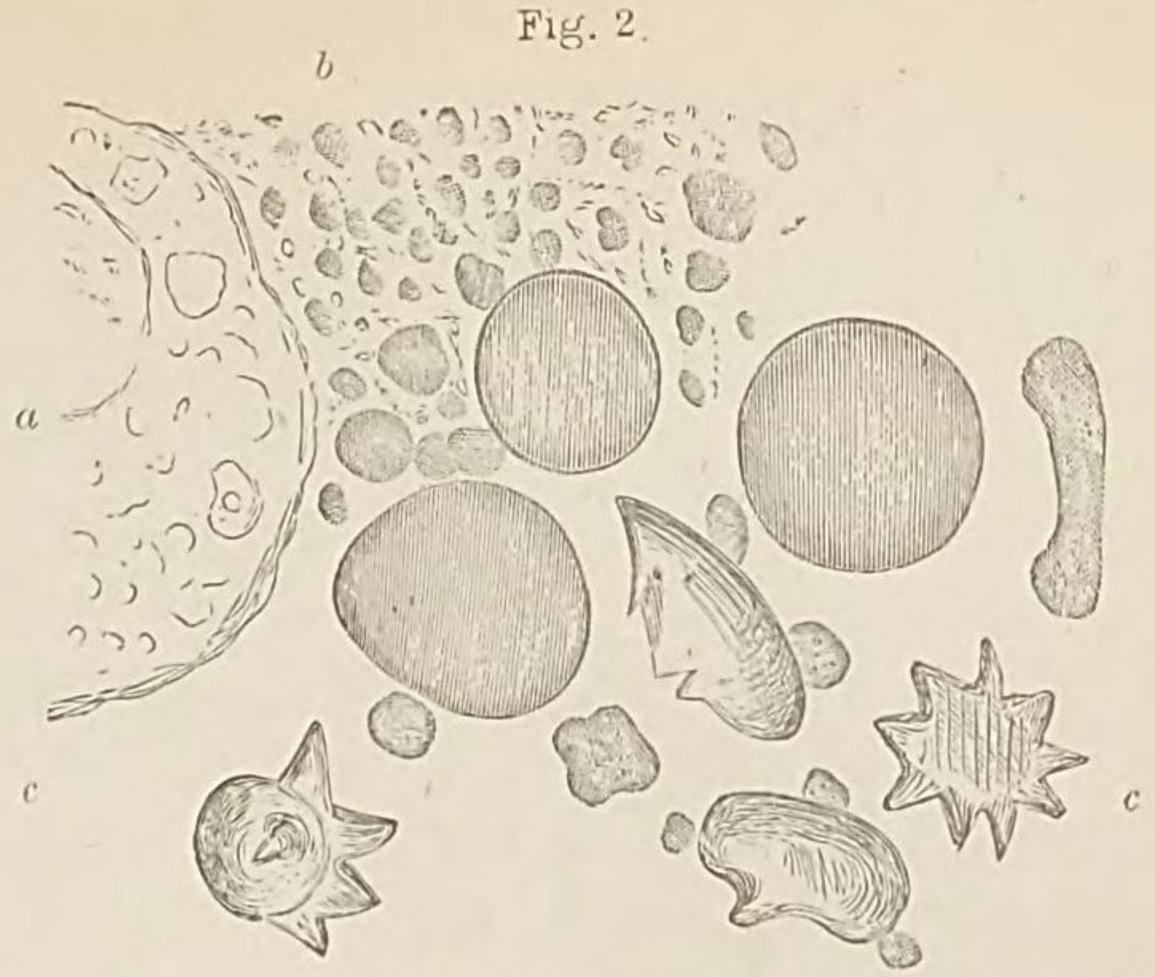
The particles of bioplasm above referred to, become obstructed in the capillaries, so that though there might be vast numbers in the blood-vessels, we should not expect to find them in the mass of the circulating fluid. Nor is it likely that they would be readily detected in blood drawn from the capillaries themselves, for they would form

Fig. 1.



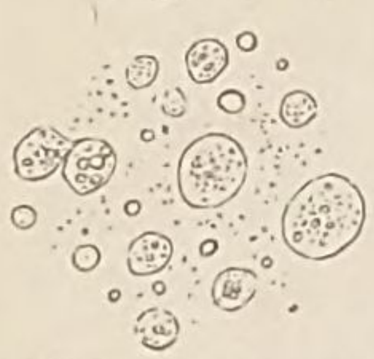
Blood corpuscles, adhering very intimately to each other, when pressed beneath the thin glass they separated, but soon became attached again. On attempting slight separation they became drawn out in a string-like form. × 215. p. 264.

Fig. 2.



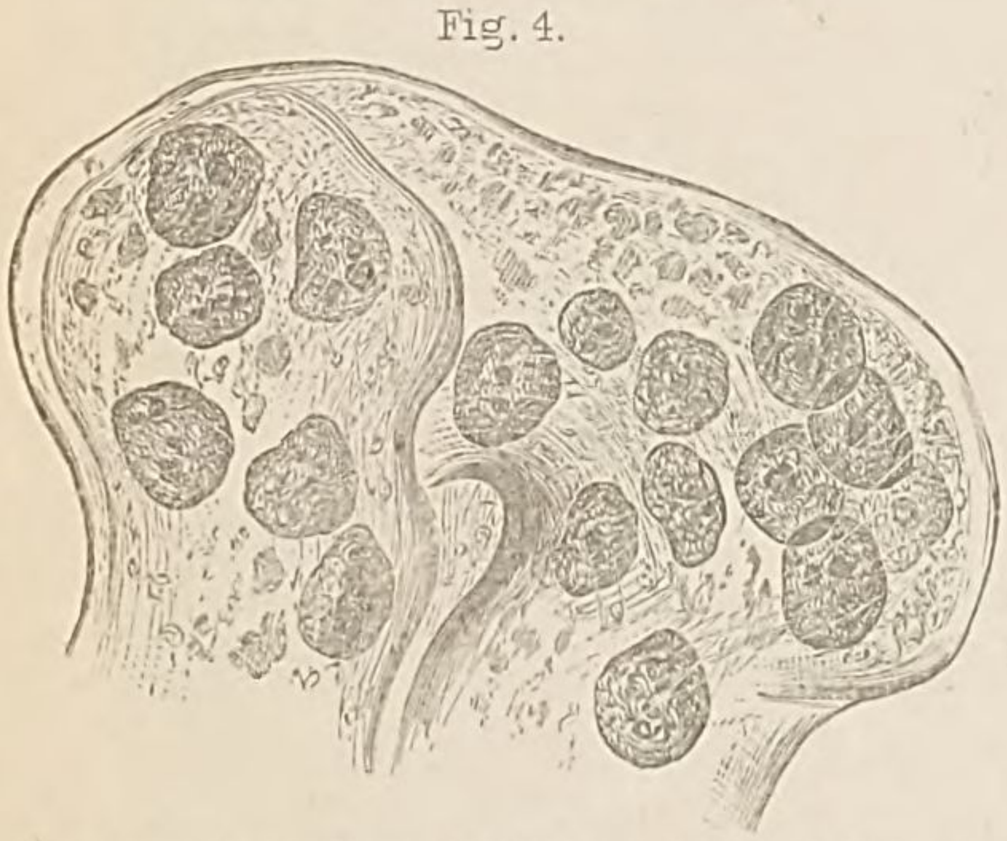
Blood pressed from intestinal capillaries into a small mesenteric vein. Cattle plague. *a*, part of white blood corpuscle in outline. *b*, minute particles of bioplasm in immense number in all parts of the field. The smooth round bodies are young red corpuscles. The angular corpuscles *c*, are old and altered red blood corpuscles. × $\frac{1}{50}$th. pp. 264, 320.

Fig. 3.



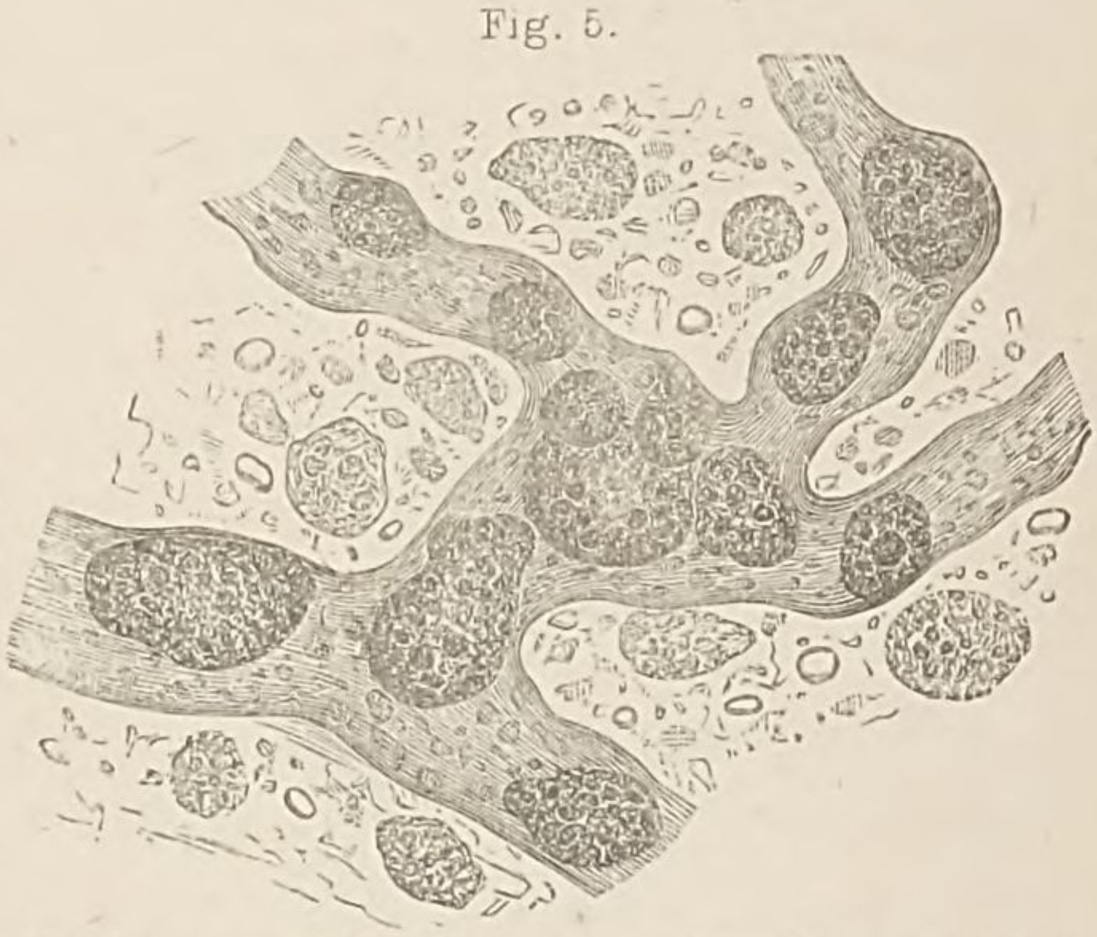
From the blood of a patient suffering from cholera. × 215. p. 263.

Fig. 4.



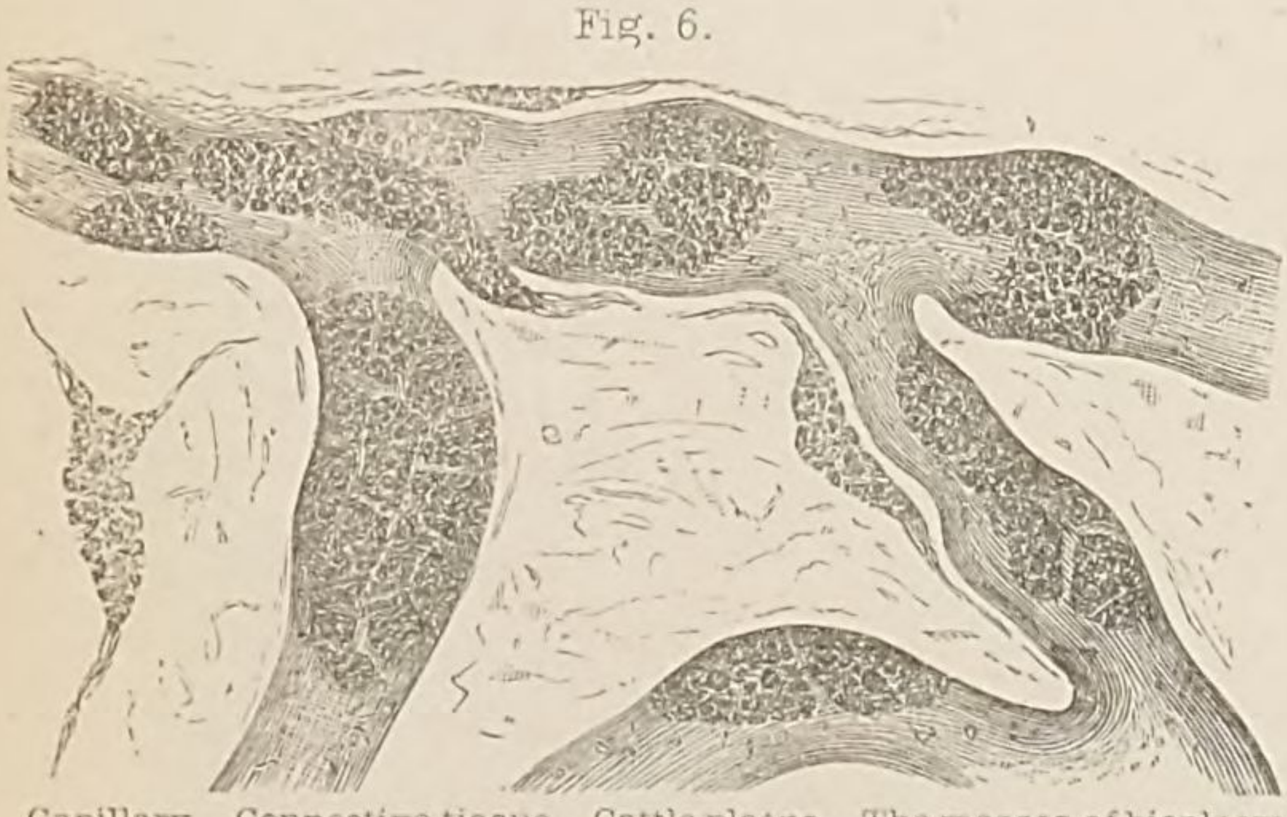
Capillary loop from Malpighian body of kidney. Cattle plague. Containing numerous white blood corpuscles and many minute particles of bioplasm (contagium). × 700. pp 261, 320

Fig. 5.



Capillary vessel from the surface of a villus, containing large masses of bioplasm. × 700. pp. 261, 320.

Fig. 6.



Capillary Connective tissue. Cattle plague. The masses of bioplasm of the capillary are very much enlarged, and are dividing and subdividing to form new masses × 700. p 246.

Fig. 7.

5

Deposit from ovarian fluid, *c*, blood corpuscles rendered spherical by endosmosis.
× 215. p. 270.

1000th of an inch ——, × 215.
" " ——————, × 700.
" " ————————————————————, × 2800.

[To face page 264

little collections which would not readily escape, but would adhere to the capillaries. From these collections particles might make their way through the walls, and then grow and multiply in the surrounding tissues. Hence in order to demonstrate these particles, I have shown that *we must most carefully examine the tissues themselves* in the manner referred to in Ch. VII. Without proper preparation, as is well known, not even the smaller vessels can be demonstrated. By squeezing the blood *from* the capillaries towards an opening in a vein, I have, however, succeeded in obtaining clots with numerous particles of bioplasm, which I believe to be the actual *materies morbi*, or contagium. The examination must be made very soon after death. By the death and decay of these particles of bioplasm numerous granules, many oil globules, and myelin particles result.

I discussed this subject in my reports on the Cattle Plague and on Cholera. It has been subsequently more fully considered in my work on "Disease Germs."

261. Examination of Blood of Lower Animals.—The blood of the lower animals, particularly of the frog, newt, and fish, should be examined when opportunities occur. The oval form of the corpuscles seems to be determined by the peculiar circumstances of the circulation in these animals, for the oval corpuscles (except the oldest, fig. 2), of the frog assume the spherical form if placed in glycerine and water, pl. XXXVI, figs. 1, 3, 4, 5. The size of the colourless corpuscles in these different animals may be compared, and it is interesting to observe the relation which they bear in size and number to the red globules.

In the substance of some of the blood corpuscles of the spleen of the dog, and of certain fish, as the perch, and other animals, two or three little yellowish crystals were observed by Funke and Kölliker. Sometimes, in examining a clot of blood, which has been effused in the brain, or in other situations, and which has remained there for some time, red crystals of hæmatine may be found in connection with altered red blood corpuscles. The subject of blood crystallization has been considered in page 191.

The phenomena of the circulation of the blood are better studied in the foot of the frog, in the tail of the minnow or stickleback, or in the branchiæ of the young newt. Among mammalia, in the wing of the bat. See page 134.

MEDICO-LEGAL INVESTIGATIONS.

262. Of Examining Blood Stains in Medico-Legal Enquiries.—In these investigations the skilled witness is often called upon to determine whether a red stain is caused by blood, and, if so, whether the blood is that of the human subject or of one of the lower animals. The latter of these enquiries is most difficult to answer, if we have to rely upon

scientific evidence alone. In some instances, although after examination we may feel pretty sure in our own minds as to the real nature of the blood, I can hardly think that in any given case the scientific evidence in favour of a particular blood stain being caused by human blood, will be of a kind that ought to be considered sufficiently conclusive to be adduced, for example, against a prisoner upon his trial. At the same time cases will occur in which a strong presumption may be of value in weakening or strengthening circumstantial evidence which is not perfectly conclusive. In all cases the medical witness should lay the facts before the jury, and clearly explain the manner in which the observations have been made upon which his opinion has been grounded, and, if he is in doubt, he should take care not to allow counsel to represent his opinion as anything but doubtful.

In testing a stain for blood we may endeavour to discover the red blood corpuscles; we may apply tests for detecting the colouring matter of the red blood corpuscles; and we may dissolve out the serum, and test the solution for albumen. The method of performing the last operation need not be discussed here, but it may be well to refer to the detection of the red blood corpuscles, and to the method of obtaining hæmin crystals, as well as to a test for blood-colouring matter, which has been considered of great value. The spectroscopic examination of blood has been already fully described (see p. 206) and is probably the method of examination least open to objection, if conducted with due care by one who has already had considerable experience. The application of the spectrum microscope to the detection of blood stains has on the whole been most satisfactory. So small a quantity can be detected with perfect certainty, under circumstances that would interfere with the success of other methods. Probably for the future the spectrum test will be that universally adopted. As already stated, Mr. Sorby has devoted a great deal of attention to this question, and an account of his method of examination has been given on p. 206 to p. 214.

On detecting Red Blood Corpuscles in a Blood Stain.—The blood stain, or a small fragment of the matter supposed to be blood clot, is moistened with a one per cent. aqueous solution of common salt. After remaining in this fluid until properly softened, the thin glass cover is applied, and the specimen subjected to microscopical examination under a power which magnifies at least 900 diameters. The blood discs will be seen very distinctly, and may be measured. See pl. XXXV, p. 258.

There are few things that can be mistaken for blood disks except the sporules of some fungi, which however are hardly ever met with in sufficient number to deceive a practised observer. As a matter of precaution, however, the observer should invariably compare the cor-

puscles supposed to be blood with actual blood corpuscles. In medico-legal investigations not only should the comparisons be made, but both slides should be kept, so that the specimens can be examined by others and compared.

If we can succeed in obtaining good specimens of the corpuscles, we may with certainty distinguish a blood stain from every other substance in nature. But in many legal enquiries this information is of little importance, unless we are at the same time able to prove that the stain in question is actually human blood, and is certainly not due to the presence of blood of one of the lower animals. This is a serious difficulty, and up to this time I fear we must admit that we are unable to decide with sufficient certainty to justify us in giving our evidence in a court of law. Though one might feel quite sure that a given specimen of blood was that of the human subject, or that of an ox, sheep, or goat, it must be borne in mind that before we give evidence we ought to be equally certain the blood could not have deen derived from the horse, pig, dog, cat, or any other animal whatever. A feeling of certainty in our own minds is however not enough. We ought to be able to bring forward evidence that would convince any unbiassed person who possessed the requisite technical knowledge and skill to enable him to form an opinion. Careful measurements of the blood corpuscles of animals have been repeatedly made. Of late years the use of very high powers, such as the $\frac{1}{25}$, has enabled us to measure individual corpuscles with great accuracy, and to determine the mean diameter in the case of any species of animal, but still there are sources of error which must not be lightly passed over. The size of the blood corpuscles varies in certain diseases, and it would not be difficult to find specimens, the mean diameter of which was above or below that of healthy blood corpuscles.

Dr. J. G. Richardson ("American Journal of the Medical Sciences," July, 1874) has very recently made some accurate measurements of the red blood corpuscles, under the $\frac{1}{25}$, and gives the following as the mean of several observations:—*Pig* $\frac{1}{4230}$, *ox* $\frac{1}{4267}$, *cat* $\frac{1}{4404}$, *horse* $\frac{1}{4600}$, *sheep* $\frac{1}{5300}$, *goat* $\frac{1}{6366}$, *human* $\frac{1}{3266}$. He therefore holds that we can, by measuring the corpuscles under the $\frac{1}{25}$, distinguish human blood stains from the blood stains of the animals enumerated above. Dr. Richardson's observations are interesting and well worthy of being carefully repeated, but I do not think that at this time anyone ought to rely upon the results of this method of investigation as sufficiently reliable for bringing under the notice of a jury.

On obtaining Hæmin Crystals from a Blood Stain.—A portion of the supposed blood clot is placed on a glass slide, and a drop of water containing a mere trace of common salt is added. The whole is then covered with a piece of thin glass, and a little glacial acetic acid is so

placed that it will gradually run in and mix with the blood. Heat is to be applied by means of a spirit lamp until the mixture almost boils. The slide is then quickly placed under the microscope, and the minute but highly characteristic rhomboidal crystals will be seen under a quarter of an inch object-glass. *See* plate XXII, fig. 7, page 194.

The Guaiacum Test.—This test, originally suggested by Van Deen, depends upon the ozone of the hæmoglobin of the blood, causing a bluish tint to appear in a properly prepared guaiacum solution. The juice of the cherry and some other fruits gives a feeble and slow reaction, but which could hardly be mistaken for that produced by blood. The tincture of guaiacum is made by dissolving one part of the resin in six parts of alcohol of 80 per cent. The bottles are to be only half filled with the tincture, so that it may be in contact with air. Strips of white blotting-paper are to be soaked in the prepared tincture, and the alcohol allowed to evaporate. If now a weak solution of blood be dropped on the paper, or applied with a glass rod, a blue colour is immediately produced. Some improvements in the use of this test have been recently recommended by Dr. Taylor. *See* also a paper by Dr. F. Falk, *Berliner Klinische Wochenschrift*, No. 49, Dec. 2, 1872, p. 590. Taylor's Medical Jurisprudence. The report of the Medico-Legal Society of Paris, "Annales d'Hygiène," July, 1873.

Serous Fluids.

263. Examination of Serous Fluids.—A serous fluid which is to be subjected to microscopical investigation should be poured into a conical glass vessel, and allowed to stand until all the deposit suspended in it has collected. A small quantity may then be removed by a pipette, in the usual way, and examined in the microscope. The microscopical characters of a serous fluid of doubtful origin, should be contrasted with those of ascitic fluid, the fluid of hydrocele, and serum from ovarian and other cysts. Portions of hydatids and claws of echinococci are sometimes met with in fluids removed from a cavity which contains, or communicates with, an hydatid cyst. The deposit should be carefully examined in the microscope, as the hooks are readily detected, and the nature of the case at once becomes evident. The deposit from a serous fluid removed from the chest of a girl, is represented in pl. XXVI, fig. 1, p. 230. Albumen in a serous fluid can always be detected by the application of heat, or upon the addition of nitric acid.

264. Fluid from Serous Cavities.—The clear serous fluid which collects sometimes to a great extent in the peritoneal cavity (ascites), will be found, if recently effused, to contain but traces of cells, or cell débris ; but after the disease has been of long standing, the surface of

the peritoneum becomes altered, and covered with a vast number of granular and almost spherical bioplasts, varying very much in size, and not usually containing a distinct nucleus. A moderately-abundant deposit often takes place after the fluid has stood for some time. In other cases, which are of a more acute character, the fluid is found to be of a greenish or dirty-yellow colour,—opaque, with numerous flocculi and shreds of false membrane suspended in it, or attached to the surface of the peritoneum. In such a specimen, pus globules, with many of the bioplasts above referred to, and fibrillated shreds of fibrin, would be found with other cells, which are darker in consequence of being filled with minute oil globules. The flocculi present a delicately fibrous appearance, with numerous cells entangled in the meshes formed by the interlacement of the fibres. Plates of cholesterine are sometimes found in ascitic fluid. The fluid which accumulates in hydrocele is usually perfectly clear, containing a few granular bioplasts, and, perhaps, a few free oil globules; spermatozoa are sometimes met with, and occasionally many plates of cholesterine are present.

Serous fluids generally contain a substance which when mixed with *globulin* forms the coagulum ordinarily known as fibrin. It was shown by Dr. Andrew Buchanan in 1845 that the fluid of hydrocele, which seemed to contain no fibrin, coagulated if a few blood corpuscles were mixed with it. The researches of Alex. Schmidt, of Dorpat, in 1861, established the important fact that for the formation of fibrin two substances were necessary, *fibrinogen* and *fibrinoplastin*. The latter is in fact globulin, or very closely allied to it. The former is contained in the fluid of hydrocele and many serous fluids, and if globulin in any form be added, *combination takes place and fibrin is formed*. Globulin is contained in the red blood corpuscles, in the colourless and lymph and chyle corpuscles. It, or some closely allied substance, exists also in the crystalline lens, in the vitreous humour, and in connective tissue. It is found in saliva, and in several other animal fluids.

265. Fluids from Cysts.—Upon contrasting the chemical and microscopical characters of the serous fluids just alluded to with those which are found within the cavities of cysts, a marked difference is always observed. As an example of a cystic fluid, ovarian serum may be instanced; but the fluid found in cysts occasionally met with in different parts of the body, as in the antrum, in the eyeball, thyroid gland, the mamma and other organs, will be found to present very similar characters. Most of these fluids contain a varying number of spherical masses of bioplasm, not unlike lymph or pus-corpuscles. Like those bodies, they are free, and multiply by forming offsets or diverticula, which become detached. Those which grow slowly are comparatively firm, while those which multiply quickly and are often present in vast numbers, are soft and easily broken down. These last

might, in certain instances, be fairly termed pus-corpuscles. In fact, the bodies in question may be regarded as slowly-growing pus-corpuscles. If inflammation be excited in the lining membrane of a cyst, the bioplasts grow more quickly, and their descendants exhibit all the characters and properties of ordinary pus-corpuscles.

The deposit of *Ovarian fluid* consists usually of cells, free granular matter, oil globules, and perhaps blood corpuscles. Not unfrequently, many crystalline plates of cholesterine are observed in it. The cells are composed of at least two distinct forms:—1. Small, delicate, transparent, and faintly granular bioplasts, without the slightest appearance of a nucleus, some being somewhat larger, and others smaller, than a pus corpuscle. 2. Large bodies, often as much as the thousandth of an inch in diameter, but varying in size, of a dark colour by transmitted, and white by reflected light. These, which have been termed "granular corpuscles," "compound granular cells," "inflammation globules," &c., are aggregations of minute oil globules in a cell form. They are almost constantly present in the fluids which are now under consideration, and have a structure apparently identical with that of bodies presenting similar characters, and frequently found in softening of the brain, pl. XXXV, fig. 11, p. 222,—sometimes in the coats of vessels undergoing fatty degeneration, in the sputum—especially in pneumonia in an advanced stage, in cystic tumours of the breast, in malignant growths, in the urine in certain cases, and in other fluids and solid structures in a state of degeneration. In all instances, the fatty matter abounds in cholesterine, which crystallizes out of the oily fat in which it was dissolved. I have seen cases in which the cholesterine crystallized after bodies of this kind had for some time been preserved as permanent objects. Its presence can always be demonstrated by treating the cells with a little dilute alcohol and allowing the solution to evaporate spontaneously. The attention of the student is particularly directed to the occurrence of bodies of this description in various morbid products.

Fig. 7, pl. XXXVII, represents the appearance of the deposit from a specimen of serum obtained from a case of ovarian dropsy. In some rare cases ciliated epithelium is met with in the fluid of ovarian cysts. Fig. 6, pl. XXX, p. 238, was taken from a specimen I met with many years ago. The cyst from which it was removed was originally developed from the ovary, and was not connected with the Fallopian tube.

CHAPTER XIV.

SALIVA, SPUTUM, VOMIT, FÆCES. — DISCHARGES FROM BOWELS, UTERUS, &c.—*Examination of Sputum.—Of Preserving Specimens of Sputum.—Extraneous Substances in Sputum.—Mucus.—Sputum in Bronchitis; Pneumonia; Phthisis; Tubercle; Fragments of Lung Tissue; Calcareous Substances.—Diphtheria.—Detachment of Flakes of Epithelium from the Mouth, Tongue, and Œsophagus.—Entozoa and Vegetable Organisms in Sputum.—Other Structures met with in Sputum.—Examination of Vomit.—Flakes of Epithelium from the Stomach.—Examination of Bile.—Examination of Matters passed by the Bowel.—Examination of Milk.—Detachment of Epithelium, and of the Examination of Discharges from the Uterus and Vagina.*

I HAVE not found it easy to arrange this part of my subject in a strictly scientific manner, but in this, as in many other cases, there is little objection to sacrifice strictness of arrangement if practical convenience and clearness are gained thereby. In this chapter I shall refer to the microscopical characters of discharges from the mouth, stomach, bowels, uterus, and vagina. In chapter XV, under the heads of *pus*, *tubercle*, and *disease germs*, I shall discuss the characters of some of the most important constituents of discharges in certain cases which have special and peculiar characteristics. Chapter XVI will be devoted to urinary deposits.

Discharges from the internal organs of the body take a great variety of forms, and he who is not familiar with the principal alterations occurring in the principal secretions and discharges in disease, will be ignorant of some of the first principles of medicine. Every secretion which leaves the body is complex, both as regards its physical and chemical composition. Departure from the healthy characters may be due to altered chemical composition, to the presence of bodies which ought not to be in it, to excess or deficiency of anatomical elements usually found, or to the abnormal characters of one or more of those elements.

SALIVA AND SPUTUM.

266. Examination of Saliva.—The examination of saliva presents no difficulty. The fluid is perfectly transparent and viscid, but it holds in suspension, besides epithelium from the mouth, a number of small

bodies, for the most part of an oval or spherical form, which are probably derived from the follicles of the gland. These are about the 1-2000th of an inch in diameter, and are sometimes called "Salivary Corpuscles." In some cases they accumulate in great number, and look like pus corpuscles, pl. XXXVIII, figs. 5, 6. A few salivary corpuscles are represented in fig. 2, pl. XXXVIII, and one or two very highly magnified are shown in fig. 4. Some observers consider them to be altered epithelium from the cavity of the mouth, but they certainly are in the saliva, and they are often met with in the absence of any of the characteristic cells of scaly epithelium, which are detached from the mucous membrane of the mouth, fig. 2. They are found in great number in some cases of salivation, and are probably the agents concerned in the conversion of starch into sugar, which invariably takes place when starch is exposed to the action of saliva. In the somewhat viscid matter of which the salivary corpuscle is composed are multitudes of somewhat highly refracting particles. These are always in active movement, and seem to be incessantly passing to and fro from the central part, where the nuclei are situated, towards the periphery of the corpuscle, and back again. The movement may be well seen under a $\frac{1}{12}$ of an inch object glass, but the observer will need a $\frac{1}{25}$ or a $\frac{1}{50}$ if he desires to study the movements of the particles accurately. A drawing under the $\frac{1}{25}$ is represented in pl. XXXVIII, fig. 4. The nature of the minute particles is extremely doubtful. They look very like the germs of bacteria, or of certain fungi—the so-called *microzymes* of recent writers, and it is possible they may be of this nature. The salivary corpuscle with its moving particles constitutes a good test object for a quarter of an inch object glass.

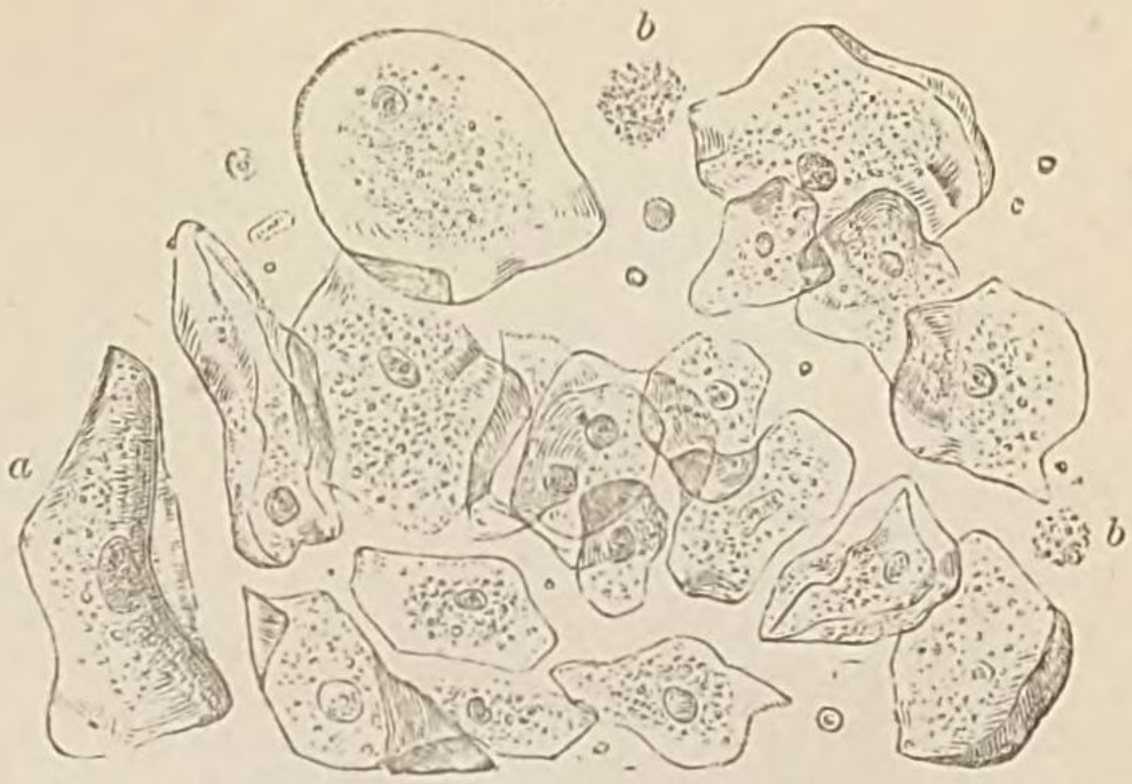
Occasionally the salivary ducts have been found to contain a considerable number of small, white, granular masses, which are perfectly spherical, and consist of cells filled with large oil globules, or the bodies themselves are perhaps mere collections of oil globules. Sometimes microscopic calculi are found. Epithelium from the mouth is represented in pl. XXXVIII, fig. 2, and in pl. XXXIX, fig. 2.

Sputum.—It is proposed to give a short description of the microscopical appearances of some of the chief varieties of sputum which come most frequently under the observation of the practitioner,—the nature of which ought to be considered in attempting to arrive at a diagnosis of the case. It is now generally admitted that in some cases much is to be learned by a careful examination of the sputum in the microscope, and there are even a few instances in which the nature of morbid changes going on during life has been ascertained, and a decided prognosis justified at a very early period of the disease, when there were really no other symptoms to attract special attention, and but little was discoverable from a most careful investigation of the

Fig. 1.

From the dorsal surface of a healthy tongue. *a*, oil globules. *b*, squamous epithelium. *c*, vegetable growths. *d*, separate filaments of fungi. × 215. p 274.

Fig. 2.

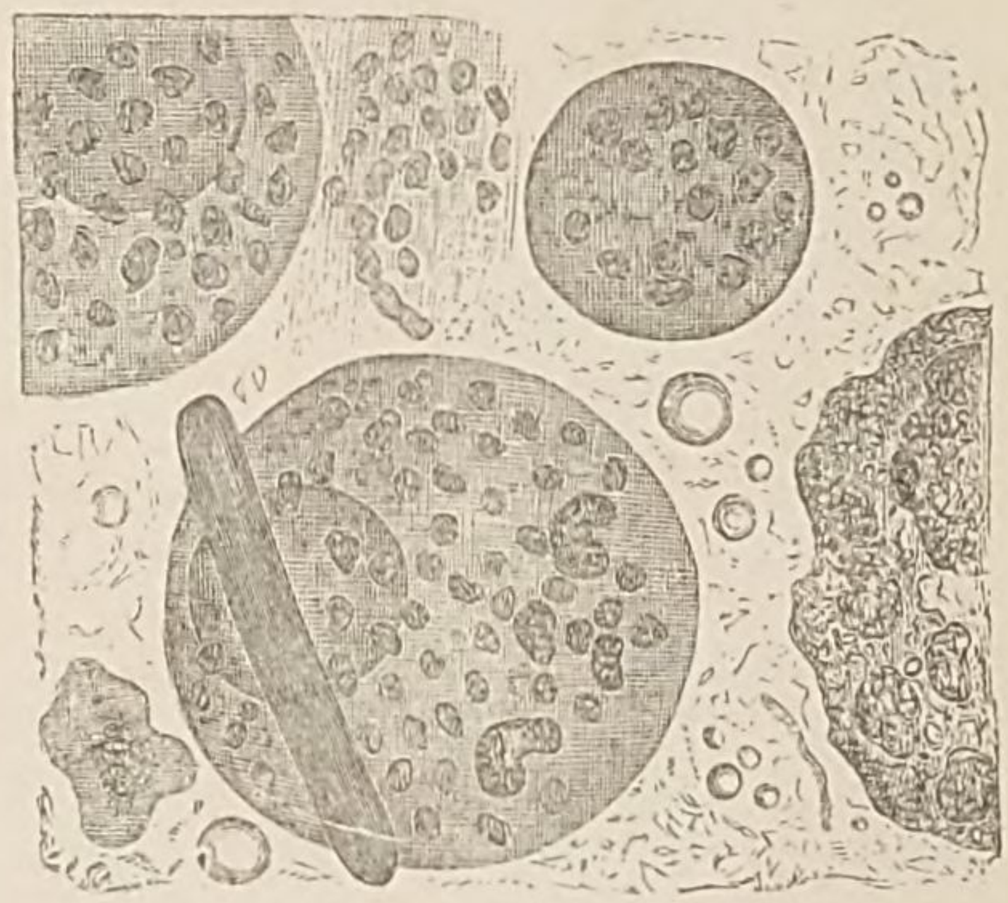


Epithelium from mucous membrane lining the cheek in health *a*, epithelial cells. *b*, sporules of fungi, the so-called microzymes p. 274.

Fig. 3.

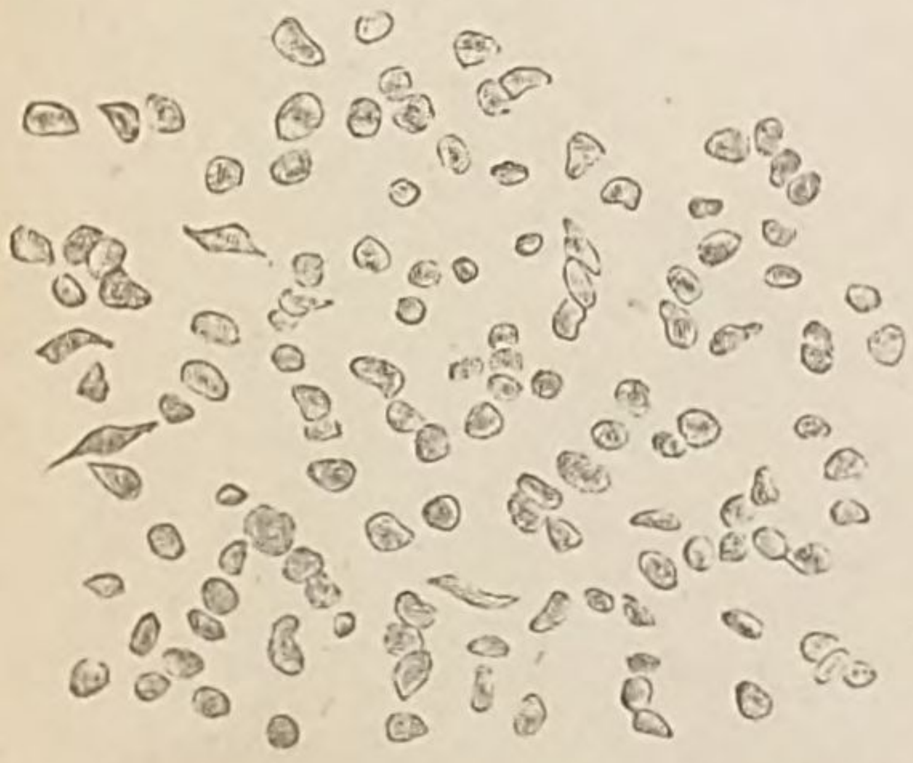
Collections of mycelia of fungi, very abundant in sputum following hæmorrhage from the lung. × 215. p 274.

Fig. 4.



Salivary corpuscles, bactrin, and bacteria germs, and oil globules from healthy saliva. × 1700. p 272.

Fig. 5.



Corpuscles which accumulated in a follicle of a salivary gland, the duct of which was obstructed. × 215. p. 272.

Fig. 6.

The same as Fig. 5, but acted upon by acetic acid. × 215. p 275.

1000th of an inch ,___, × 215.

[To face page 272.

physical signs. It is, however, quite true that the nature of many cases is to be satisfactorily ascertained without resorting to a microscopical examination of the sputum, and in some cases the microscope does not afford any help in diagnosis. Every practitioner should, however, be familiar with the microscopical characters of the principal varieties of sputum; for in the course of practice he will certainly meet with obscure cases, in the diagnosis of which the microscope will afford him valuable aid. The observer should also be acquainted with the different forms of epithelium which occur in sputum, particularly the epithelium from the mouth and tongue, and he should remember that many small particles of food are often found entangled amongst the long hair-like epithelial growths detached from the conical or fungiform papillæ, and may be mixed with the sputum.

In searching for any particular substance in sputum, we must not rest contented with the examination of one, two, or three specimens; but many portions, taken from different parts of the mass, must be successively examined. When fragments of pulmonary tissue are to be sought for, the examination should be conducted with great care, and several specimens should be placed one after the other under the microscope, before any conclusion is adopted. Small particles are often scattered sparingly through the mass, and may thus easily escape observation. *See* page 279.

267. Examination of Sputum.—Some observers have recommended that sputum, intended for microscopical examination, should be thrown into water, so that certain pieces may be selected; but I think, as a general rule, it is better to avoid the admixture of water, as a physical alteration is produced in many of the cells, and the complete disintegration of some is effected. Small pieces of sputum should be removed from the vessel with the aid of forceps and scissors, and placed upon a glass slide. Two or three specimens from different parts of the sputum may be removed at once, and placed on the same glass slip for examination. As some difficulty is often experienced in consequence of the tenacious character of the sputum, Dr. Sansom designed a pair of forceps which to some extent overcome this difficulty. These are represented in pl. VI, fig. 8, p. 74. The blades are slightly cup-shaped and the edges sharp, so that pieces of the viscid sputum can be easily cut off. Pieces of sputum will often require to be teased out with needles upon the slide, and if, from the opacity of the specimen it is necessary to add a fluid, it is better to use a little glycerine and water, or white of egg instead of pure water. The specimen to be covered with thin glass in the usual manner.

268. Of preserving Specimens of Sputum permanently for Microscopical Examination.—Specimens of sputum may be preserved in glycerine and water, in which fluid they keep very well, but are

rendered very transparent; the naphtha and creosote solution, dilute spirit, and water impregnated with arsenious acid are also employed for preserving sputum. The preservation of the recent characters of sputum is a matter of great difficulty. I have tried a great number of different preservative solutions, but have not succeeded in finding one which possesses all the qualities required. Many so completely alter the character of the anatomical elements, that they could not be recognized, while some have the effect of keeping the specimen very well for a time, but after the lapse of some months it will be found to have undergone complete change. Upon the whole I find glycerine the best basis for a preservative medium; but inasmuch as strong glycerine renders many specimens of sputum too transparent, glycerine and camphor water, or glycerine and water with a trace of carbolic acid, are to be preferred.

269. Epithelium and Extraneous Substances in Sputum.—Epithelium from the cavity of the mouth and air passages, with portions of any vegetable growths as leptothrix, fungi, the so-called microzymes, and fully developed bacteria, which are so commonly found in the mouth, pl. XXXVIII, figs. 1 and 3, especially upon the dorsum of the tongue and amongst the matter secreted by the tonsils, with small fragments of any substances taken as food, may be met with in sputum. Unless the observer is familiar with the appearance of all these bodies, he will find himself beset with difficulties at every step, and will be liable to fall into the most ludicrous errors.

In the first instance, he should make himself familiar with the characters of the epithelium from the cavities of the mouth, pl. XXXVIII, fig. 2, nose, tongue, trachea, and bronchial tubes, and with the bioplasts in the mucus formed upon these portions of the mucous membrane, p. 229. Next, he should place under the microscope small quantities of the different extraneous matters likely to be met with most frequently. The most important are the following: bread, wheat starch, potato starch, rice starch, testa of wheat, cells of potato, and other vegetables taken as food, cotton, flax, and silk fibres, portions of feathers and hair, air bubbles, oil globules, portions of adipose tissue, as bacon, muscular fibre, white and yellow fibrous tissue, fragments of cartilage, bone, &c. Some of these are figured in pl. XLI, others will be found in pl. XLVII, containing extraneous matters often detected in urine.

Of the Different Kinds of Sputum.

The characters of sputum are much influenced by the time which elapses between its formation and its expectoration; and as already mentioned its anatomical elements are numerous and variable.

Sputum often contains mucus corpuscles from the mucous mem-

brane of the nasal, tracheal, or bronchial passages. These may easily be mistaken for colourless blood corpuscles, and have in fact been spoken of as leucocytes, pl. XXXVIII, figs. 4, 5, 6. The observer, who has already made himself familiar with the characters of the "mucus" of the respiratory passages, will at once recognise the bodies in question. No one should consider himself properly qualified to enter upon microscopical investigations in connection with disease until he has been through a course of preliminary study.

The characters of the "mucus" vary much, and it is desirable to direct the reader's attention in this place to the important alterations which take place in the characters of mucus as it passes from the usual (*normal* would scarcely be accurate) into the morbid condition.

270. Mucus, which is formed upon the fauces, and upon the mucous membrane of the nose and air tubes of healthy persons is clear and transparent. The viscid, indistinctly fibrillated material, to which the physical characters of the "mucus" are due, entangles in its meshes cells of various forms and in different stages of growth; and in some specimens every transitional form of cell, from the large cell of squamous epithelium to the small faintly granular corpuscle, formerly termed *mucus corpuscle*, may be detected (see pl. XXVI, figs. 8, 11). Not unfrequently cells of columnar ciliated epithelium from the trachea or bronchial tubes are present. Fig. 3, pl. XL, represents the microscopical characters of a specimen of transparent, frothy, viscid, and almost colourless bronchial sputum. Some of the cells which have been treated with acetic acid are shown to the right of the figure. Clear mucus is coagulated by acetic acid, and numerous striæ make their appearance.

The little oval particles, seen embedded in the transparent slightly streaky viscid mucus, consist of bioplasm. In an ordinary "cold" these increase in size and multiply in number, in consequence of the supply of pabulum, which transudes from the blood, being more abundant than in the perfectly normal or healthy state. Probably a departure from the normal state has taken place in the blood before the change in the mucus corpuscles occurs. This consists of the production of an undue proportion of soluble organic materials,—certain of the so-called extractives, which readily permeate the capillary walls and would undergo chemical change, and do harm were they not appropriated by bioplasm particles. The mucus bioplasts not being invested with a layer of firm formed material (cell-wall) are among the first to appropriate the excess of soluble organic material referred to. Accordingly, the bioplasts grow, and give off diverticula, which become detached and grow. The process may continue until the "mucus" which is secreted appears yellowish (muco-pus, purulent sputum) from the great number of bioplasts it contains. When examined in the microscope this mucus is

seen to consist almost entirely of these particles of living bioplasm separated by, or imbedded in a very small proportion of the "mucus," or formed material to which the tenacity of the ordinary secretion is due. The changes which take place in the bioplasts and epithelial particles of the mucous membrane of the fauces are represented in pl. XXXIX, figs. 1 and 2. The latter figure was copied from some young epithelial particles which were changing at the time. They were growing much more rapidly than in the normal state, in consequence of the supply of pabulum being increased. Such rapidly formed anatomical elements would form a very soft spongy epithelial layer easily detached in the form of shreds of mucus, which, when removed, would be soon replaced by others of the same kind. Such changes occur commonly enough upon the mucous membrane of the upper part of the pharynx.

In *catarrh*, when the "mucus" is more abundant, besides the mucus and pus bioplasts above alluded to, a number of round or oval masses are observed, which consist of aggregations of minute oil globules cohering together, and often appearing as if they were within a cell wall. Two or three of these are represented in fig. 3, pl. XL. They are often present in great number. Granular masses, varying much in size, but for the most part smaller than the last, are also met with. A vast number of granular corpuscles, closely resembling pus corpuscles, are very common in most specimens of sputum. It is not difficult to make out the intermediate stages between the faintly granular particle which is rendered transparent upon the addition of acetic acid, and exhibits a nucleus, and the true pus corpuscle, in which this reagent develops two or three highly refracting bodies, a circumstance which distinguishes pus from different forms of young epithelial particles.

271. Sputum in Pneumonia.—In the rust-coloured sputum of the early stages of acute pneumonia will be discerned a number of the large spherical collections of minute oil globules which used to be called exudation corpuscles, or granular cells, together with a vast number of minute granular particles of bioplasm of a circular form which grow and multiply in the matter which occupies the air cells of the lung, besides numerous blood corpuscles, for the most part separated from each other. The peculiar yellowish or rust colour of the sputum is due to the circumstance that the bood corpuscles escape very gradually, as it were one by one, and thus become intimately mixed with the other constituents. At a later stage, in bad cases, the quantity of blood increases, the matter expectorated is nearly fluid, and contains a vast number of disintegrated cells and granular matter, with numerous altered and ragged blood corpuscles.

The enormous number of bioplasts present in the air cells of inflamed lungs are not colourless blood corpusles that have escaped from the vessels, but result from the growth and multiplication of

PLATE XXXIX.

Fig. 1.

Cells contained in mucus, removed from the mucous membrane of the fauces of a man suffering from relaxed sore throat. The affection was very slight, and the specimen was removed on the first day. *a*, epithelial cells. *b*, small cells containing granular matter and oil globules, consisting partly of altered epithelium, partly of mere aggregations of oil globules surrounded with viscid matter, giving the appearance of a cell wall. A group of these are represented on the right (*c*), more highly magnified. × 403. p. 276.

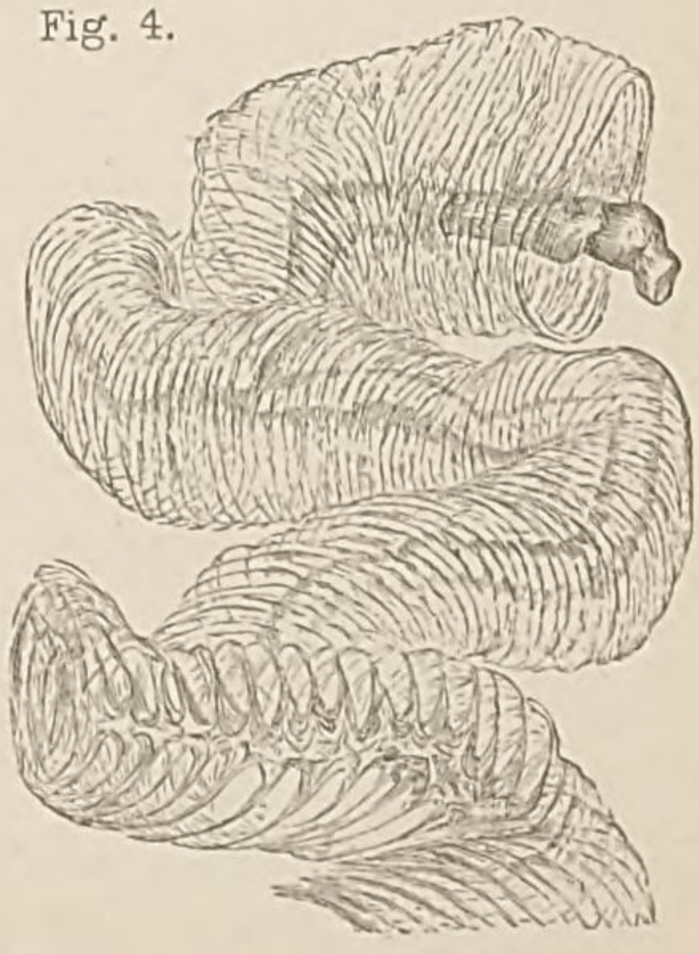
Fig. 2.

Epithelium from the surface of the pharynx of a healthy man suffering from very slight sore throat, a day after catching cold. *a*, a normal full grown epithelial cell for contrasting with the other specimens, in which the proportion of the bioplasm is very great, while the formed material is still plastic, in consequence of still having disseminated through it a good deal of untransformed bioplasm. The state depicted depends upon very rapid growth having taken place. × 700 p. 276.

Fig. 3.

A mass of lung tissue in sputum of phthisis. Some of the air spaces and smaller bronchial tubes are filled with chalk-like casts. × 215. p. 279.

Fig. 4.



The remains of a small bronchial tube, from the same sputum as represented in fig 3. × 215.

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particles of bioplasm which have passed through the stretched capillary walls, as I pointed out before 1860. See page 259. The minute particles of bioplasm suspended in the liquor sanguinis readily pass through the capillary walls, which are stretched to so great an extent that longitudinal fissures result, which indeed are wide enough to permit red blood corpuscles to pass through them. The particles of bioplasm having reached the air cell, grow, and soon give rise to the multitude of spherical bioplasts of which the "exudation" seems to be mainly composed.

The sputum in pneumonia contains a considerable quantity of chloride of sodium, which appears to be attracted as it were from the blood and from the tissues to the seat of inflammation. It is remarkable that at this time the urine contains no salt, or but mere traces, and if salt be given during the persistence of the inflammation, it will not pass away in the urine as it does in health. As soon, however, as the inflammation is resolved, the salt is re-absorbed from the products in the lung, and may again be detected in the urine.

272. Sputum in Bronchitis.—The opaque yellow sputum of chronic bronchitis owes its peculiarities to the presence of pus corpuscles which are suspended in the viscid material. In these cases many forms of corpuscles are met with, and epithelium in all stages of growth can be often discerned. Granular matter and small oil globules are frequently present in considerable numbers. Collections of dark colouring matter, more or less globular, and much resembling the collections of minute oil globules alluded to in the last section are frequently observed. These are composed partly of blacks, or particles of soot, introduced in respiration. Sometimes dark colouring matter is actually formed in the air cells of the lung, the dark coloured material being derived from the blood, and not introduced from without. A large quantity of coal dust is found in the expectoration of men working in coal mines; and in the case of the Sheffield dry grinders, metallic particles, which are inhaled, give rise to great irritation, and, not unfrequently, to death at an early age. These metallic particles are expectorated, and can be detected in the sputum. Dr. Hall, of Sheffield, has paid great attention to this fatal disease. *See* his work "On the Pathology, Diagnosis, &c., of Thoracic Consumption," third edition; Longmans, 1856. Any insoluble matter in a state of minute division which floats upon the surface of or is suspended in a fluid in motion, will tend to aggregate to form small collections; but it is not less unreasonable to assume that all things resembling these aggregations must be produced in precisely the same way, than it would be to maintain that by such aggregations is formed every anatomical element of all living beings, and that therefore no absolute line of demarcation separates living beings from lifeless mechanisms, voltaic batteries, and laboratories.

The character of the pus corpuscles varies much in different specimens of sputum. Sometimes they are well formed, and exhibit their ordinary characters, but oftentimes they are very faint, not perfectly circular, perhaps with very irregular outlines, and not unfrequently partly disintegrated. In cases where the pus has been retained for some time in the air tubes, or in cavities after its formation, it is completely broken down, and no distinct corpuscles are to be distinguished.

273. Sputum in Phthisis.—The characters of sputum in phthisis vary according to the particular form of the disease and the stage it may have reached, the particular tissues of the lung implicated, and the length of time the sputum may have been retained in the cavity before its expectoration has occurred, and many other circumstances. No physician would attempt to diagnose a case of lung disease from the examination of the sputum only; nor are the characters of the sputum so invariable as to enable us to determine with precision the particular stage of the disease. The forms of sputum which would be considered by ordinary examination with the unaided eye to be characteristic of phthisis, are met with only in confirmed cases, where there is almost invariably conclusive evidence of a different kind indicating the existence of the disease. The sputum often contains pus corpuscles, sometimes well formed, and in other instances apparently disintegrated, with much granular matter, and often minute oil globules, with a number of mucus corpuscles or bioplasts, derived from the smaller bronchial tubes. In many cases, the microscope undoubtedly affords very important information.

The general appearance of tubercle corpuscles under low magnifying powers is represented in pl. XLVI. They are seldom found in sputum unless mixed with a considerable quantity of granular matter, and can hardly be recognised with certainty. In many cases, pus corpuscles are so numerous that it is difficult to discover the tubercle, which, moreover, is often disintegrated, so as to be indistinguishable as a special element of the sputum. The nature and characters of tubercle are discussed in Chapter XV.

Fragments of Lung Tissue.—It is most important that the practitioner should be familiar with the characters of one structure, which is not unfrequently met with in phthisical sputum, and which has already been cursorily alluded to. The recognition of this is really a matter of great practical importance. The microscopical characters of lung tissue are distinct, and, by a qualified observer, the fibres can hardly be mistaken for anything else met with in sputum. The diagnosis to which the practitioner will be led even at an early period of the illness, before the patient or his friends have the slightest suspicion of serious disease, will be in almost every case in which the structure is observed, subsequently confirmed by unmistakable evidence. Professor Schroeder van

der Kolk was, I believe, the first observer who drew attention to the importance of the presence in the sputum of the elastic fibres of pulmonary tissue at a period long anterior to the development of any definite symptoms. This observer wrote in the year 1846, and the value of his observation has since been amply confirmed by Dr. Theophilus Thompson, Dr. Hughes Bennett of Edinburgh, Dr. Andrew Clarke, Professor Quekett, Dr. Fenwick, and myself, and more recently by numerous observers here and on the continent. The elastic tissue is not prone to change. Lung tissue can be detected with great facility, especially if the sputum be treated with acetic acid, which renders the other elements transparent, while it exerts no action upon the elastic tissue.

The presence of lung tissue shows that disintegration of some of the air vesicles has actually commenced. In searching for this substance, several specimens from different parts of the sputum should be examined, and any little grayish masses should be particularly selected. Dr. Bennett mentions a case in which this elastic tissue was met with at a time when no other signs of phthisis were present. The sputum was examined by Dr. Bennett, Dr. Iliff, Professor Quekett, Mr. Rainey, and myself. All concurred in pronouncing the substance to be pulmonary tissue. After a time other symptoms of the affection manifested themselves, the physical signs of a cavity became distinct, and the patient died. The lung tissue represented in pl. XL, fig. 1, was found in the sputum of a case of phthisis of about a year's duration. Figs. 2, 4, and 5 are copies of fragments of pulmonary tissue found in sputum which contained a very large quantity. The proportion of expectoration in this case was very small, amounting to not more than half a dozen pellets in twenty-four hours. The case was that of a stout lady of about fifty years of age, who had been suffering from cough, for about six weeks, consequent upon taking cold. There was slight dulness under one clavicle, but no marked symptoms of phthisis, in fact it was difficult to persuade the patient that there was anything the matter with her, and the diagnosis rested almost entirely on the fact of the presence of the pulmonary elastic tissue in the pellet of sputum which was subjected to examination.

Lung Tissue and Altered Bronchial Tubes in the Sputum of a case of Phthisis.—The interesting specimens which are represented in pl. XXXIX, figs. 3 and 4, were obtained from the sputum of a female patient, age 45, who had had cough for five or six years, accompanied by very slight expectoration. She was emaciated, but did not seem in the last stage of the disease when I saw her. The symptoms were not more urgent than they had been for a long time past, and though it did not appear probable that any improvement would take place, there was nothing in the patient's general state to be urged against the opinion that she might live for many months. *Percussion under each clavicle was*

resonant, and the expansion natural. There was, however, marked dulness in the right supra-spinous fossa, and crepitation was distinctly audible in this situation.

The expectoration at this time was very slight, but the microscope revealed a degree of destruction of lung tissue, which I was not prepared to find, and which I have never met with before, or since. It was evident that in this case the lung tissue was being rapidly disintegrated bit by bit, while very little, if any, inflammation accompanied the process. There were to be detected scarcely any of those inflammatory products, altered mucus, pus, "inflammatory corpuscles," &c., which usually constitute the great bulk of the sputum in ordinary cases of phthisis; while the small quantity of sputum formed consisted almost wholly of the structures entering into the formation of the lung tissue. In fig. 3, portions of three or four air cells of the lung are represented. The pulmonary tissue is well seen, and occupying the cavities of one or two air cells, and passing into contiguous ones, are some cylindrical masses of calcareous matter, such as are not unfrequently expectorated in some forms of phthisis. To the right of *a* is an elongated mass of altered blood, in which are observed two well-marked but minute crystals of hæmatoidin. Fig. 4 represents what is probably a small portion of a minute bronchial tube, or very much changed portion of artery, the walls of which have become much thickened and altered by slow change, and the formation of much fibrous material around it. Its narrow tube is occupied by a cylinder of calcareous deposit.

The patient died only three weeks after I saw her. Unfortunately I had but one opportunity of examining her, and there was no *post mortem*. There had been no hæmoptysis, and it appears that death resulted simply from exhaustion, which came on very rapidly shortly after she came under my observation. It seems to me probable that in this case a very slow wasting and disintegration of lung tissue had been going on for a length of time, probably for years, in several different spots separated by portions of perfectly sound lung tissue.

In order to obtain fragments of lung tissue from sputum, Dr. Fenwick recommends that the sputum be liquefied by being boiled in a solution of soda, Med. Chir. Soc., June 26, 1866. *See* also "The Student's Guide to Medical Diagnosis," by Dr. Fenwick. The following is the plan of proceeding:—About twenty grains of caustic soda are to be dissolved in an ounce of distilled water. All the sputa expectorated in twenty-four hours are to be mixed with an equal bulk of the soda solution. The whole is to be boiled in a beaker, and then four or five times the quantity of cold distilled water added. Any lung tissue will have resisted the action of the soda, and will soon subside to the bottom of the vessel. The sediment may be transferred to a conical glass, and after time has been allowed for the subsidence of solid

particles, may be removed to the glass slide with the aid of a pipette, and examined in the usual manner under a quarter of an inch object glass, after being covered with thin glass.

In some specimens of sputum there are numerous curved bands and streaks of mucus which somewhat resemble the elastic tissue; upon the addition of acetic acid no distinct fibres are to be made out, and the fibrillated appearance becomes less defined in consequence of the mucus shrinking from the action of the acid. The observer should not trust entirely to appearances, until he has made himself familiar with the characters of the elastic tissue taken from the lung itself. Crystals of cholesterine are occasionally found in phthisical sputum, and granules of phosphate of lime are also met with.

274. Entozoa and Vegetable Organisms in Sputum.—Hydatids are sometimes expectorated in sputum. Occasionally they are developed in the lung itself; but in the great majority of instances they are formed in the liver,—an opening is gradually made through the diaphragm, and the hydatids at length make their way through the lung into a large bronchial tube. After the cyst has been completely emptied, the large wound gradually closes, and the patient may get quite well. Two or three cases of this kind have been in King's College hospital. One occurred, some time since, in the practice of Dr. Todd,* and I have had two or three under my own care more recently, all which terminated in recovery. We shall probably see very few of these cases now, because we are able to operate and remove the hydatids from the liver, in many cases in which even ten years ago we should not have considered it right to interfere. The characters of the cysts are sufficiently distinctive as a general rule; but if the sputum be well agitated with water and allowed to stand, the hooklets of the echinococci will sink to the bottom and may be removed with a pipette. The appearance of these is characteristic. See drawings in plates illustrating the characters of Entozoa. In many of these cases biliary acids may be detected in the sputum, and in a case under my care a short time ago, crystals of cholepyrrhin were found in large number.

Fungi are from time to time met with in sputum (fig. 3, pl. XXXVIII), but the distinctive characters of these will be briefly considered in the last chapter. Fig. 9, pl. XL, represents the characters of fungi from some apthous sores in the mouth of a patient in the last stage of phthisis. The specimen was sent to me by my friend Dr. Scott Alison.

275. Blood Corpuscles and other Bodies met with in Sputum.—*Blood Corpuscles* are occasionally met with in small numbers in all varieties of sputum.

Blood corpuscles in sputum may come from the lung, or may be

* "Medical Times and Gazette," 1852. See also Livois, "Recherches sur les Echinocoques chez l'homme," &c.—Thèse, Paris, 1843.

derived from the gums or tongue, or they may escape from the tonsils fauces or back of the throat. Occasionally hæmorrhage takes place from the pulmonary capillaries without any serious disease either of the lung tissue or of the vessels, and, as is well known, may be a consequence of disturbed uterine action. In pneumonia, bronchitis, diphtheria, and other acute affections, blood may be present in sputum, and in some cases of aneurism there is an escape of a little blood from time to time long before the period of dangerous or fatal hæmorrhage arrives, and perhaps before it is possible to detect any dilatation of the artery by physical signs. Hæmorrhage from one or more vessels of a cavity formed in the course of phthisis is however by far the most frequent cause of blood in sputum. Wherever, therefore, blood is frequently observed, the sputum should be carefully examined for the purpose of detecting portions of pulmonary tissue, and the most careful physical examination of the chest should be made and repeated at short intervals. The student must remember that sporules of fungi are very often found in sputum, and without great care may be easily mistaken for blood corpuscles.

Dark Granular, Cell-like Bodies.—In sputum in various conditions a number of dark cell-like masses are often found. In some cases the dark material consists merely of carbonaceous matter which has been inhaled; but in other instances it seems to be composed of a dark pigmentary material derived from some portion of the respiratory tract. This substance is doubtless formed from the blood, as it is found in various organs quite unconnected with respiration. It is exceedingly common in many of the lymphatic glands, especially in those near the bronchial tubes, and has been described by some observers under the head of *melanosis;* but there are many instances in which this dark material is deposited unconnected with cancer, and these have been included under the term "*spurious melanosis.*"

Myelin and cells containing minute particles of this substance are not unfrequently found in sputum. A good illustration of this, met with in a case of hæmoptysis, is represented in fig. 8, pl. XL.

Calcareous Masses.—In a few cases of chronic phthisis, gritty masses consisting of phosphate and carbonate of lime are sometimes expectorated. These are not unfrequently as large as a pea and even larger. They result from the disintegration of tubercle, the organic portion of which has been removed by absorption. I have known them to be coughed up in several cases, and think that their expectoration is generally indicative of a favourable change. Indeed, the formation of these bodies seldom occurs until active mischief has subsided, but it is, of course, quite possible that in some other part of the lung new disease may be developed. In fig. 3, pl. XXXIX, a specimen of sputum, containing calcareous masses, is represented. It is not un-

common to meet with them in post-mortems, inclosed in a small fibrous cyst, surrounded by healthy lung.

Fibrinous casts of the large and small bronchial tubes are expectorated in certain cases, of which instances are recorded in all standard works on Medicine. Under the microscope they are seen to be composed of a striated material like fibrin with a number of small faintly granular corpuscles.

The following references will be useful to those who desire to make a special study of the microscopical characters of sputum :—

Wright, "The Pathology of Expectoration."—(Med. Times, 1844-45.)

Lebert, "Traité de la Phthisie," second edition, Paris, 1843.

Remak, "Diagnostische und Pathogenetische Untersuchungen," Berlin, 1845. Deutsche Klinik, Sitzungsprotokoll der Gesellschaft für Wissenschaftl. Medicin in Berlin, vom 1 Juli, 1850.

Shroeder Van der Kolk, "Nederlandsch Lancet," 1846. "Sur la présence des Fibres Elastiques dans les Crachats des Pthisiques," Bruxelles, 1850. "On the Origin and Formation of Tubercles in the Lungs."—(Nederlandsch Lancet, 3rd series, No. I en II.)

Hœfle, "Chemie and Mikroscop am Krankenbette." Erlangen.

Jacubowitsch, de Saliva, diss. Dorpat, 1848.

Virchow, "Verhandlungen der Physikal. Medicin. Gesellschaft in Würzburg," 2 Bd., Sitzung vom 4 Jan., 1851.

Dr. Black, "Association Journal," 1853.

Thierfelder, über Bronchitis crouposa, Archiv für Physiol. Heilkunde, 13 Jahrgang, 2 Heft. 1854.

Dr. H. Thompson, Lettsomian Lectures, 1854, "Lancet," Feb. 1857.

Dr. Andrew Clarke, in "Transactions of the Pathological Society," vol. vi, page 74.

Dr. Hughes Bennett, in "Edinburgh Monthly Journal," January, 1856, page 585. "Clinical Lectures on the Principles and Practice of Medicine," 1858.

Dr. J. C. Hall, "Hints on the Pathology, Diagnosis, Prevention and Treatment of Thoracic Consumption."—Longman, 1858.

Dr. Radclyffe Hall, "Medico-Chirurgical Review," vol. xv, page 477; vol. xvi, page 465; vol. xvii, page 449.

Dr. R. P. Cotton, "Fothergillian Prize Essay."

Dr. Th. Williams, Article "Rèspiration."—(Cyclopædia of Anatomy and Physiology.)

Dr. Anton Biermer, "Die Lehre vom Auswurf," Würzburg, 1855.

Dr. Fenwick's paper in the Med. Chir. Trans., 1866; and his Student's Guide to Medical Diagnosis."

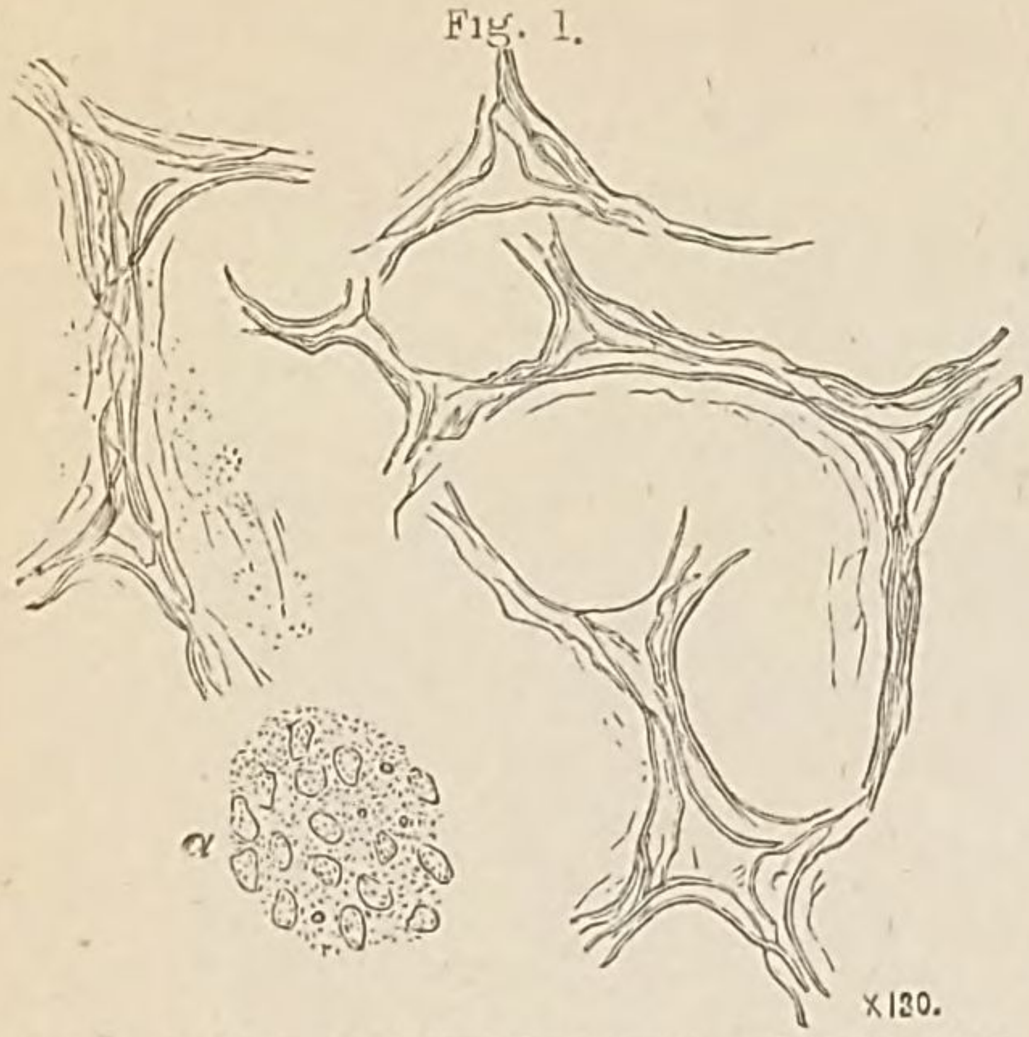
Diphtheria.

276. Diphtheria.—There is no special character by which the exudation effused upon the surface of the mucous membrane of the fauces in cases of diphtheria can be invariably distinguished. It consists, as is well known, of a white soft membrane varying considerably in thickness. Under the microscope, this is found to be composed of a more or less transparent viscid substance, about the consistence of very firm mucus, and exhibiting the striations and wavy lines always seen in this material. Sometimes the lines are so regular as to give to the specimen a delicately fibrous appearance. Entangled in this are found *a*, cells of scaly epithelium from the mouth; *b*, a number of small transparent granular, round, or oval particles of bioplasm, resembling those found in the mucous follicles of the fauces and in the deepest layers of epithelium.

In some cases, however, the membrane appears to consist almost entirely of ordinary epithelium, in others the small roundish cells predominate, while sometimes the mass appears very transparent and only contains a few of both forms of cells just described. The small bioplasts pass into pus corpuscles, and where the case is severe and the powers of the patient much reduced, the number of these pus-like bioplasts is very great. It is, however, important to observe, that the action of acetic acid upon these differs from its action upon well-formed pus corpuscles. One or two bodies with a well-defined dark outline, but not perfectly circular, are certainly displayed, as in the case of the pus corpuscle, but the greater part of the bioplasm particle seems to be dissolved by the acid, or rendered so very transparent as to be quite invisible. It is, however, probable that if the production of such particles continued for a certain period of time, well-defined pus corpuscles would at last be produced.

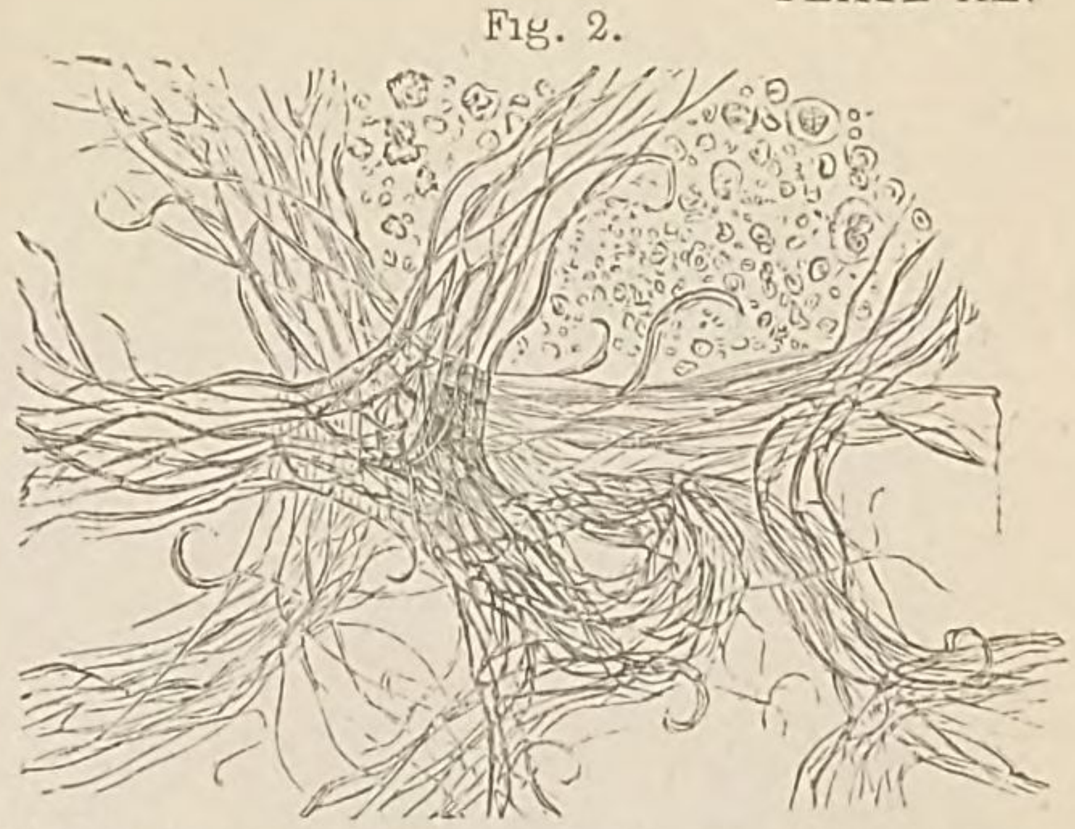
The greatest confusion has resulted from applying the same term, *diphtheria*, to several different pathological changes. It is, I think, incorrect to call a slough upon the throat "diphtheritic membrane," for the two things are pathologically distinct. Again, it is said that true diphtheria tends to result in laryngeal croup, but many practitioners have seen numerous cases of diphtheria without any laryngeal affection at all, and cases of croup, in which there was not any tendency to the formation of false membrane upon the fauces, or to the development of that soft œdematous condition of mucous membrane present in true diphtheria. The state of system which precedes the accession of diphtheria is not by any means the same as that which exists before the development of croup, and the blood is modified in diverse ways in the two diseases. In diphtheria a material, perhaps previously existing in the blood, exudes through the capillary walls, infiltrates the tissues, and escaping from the follicles, coagulates layer after layer upon the surface of the mucous

Fig. 1.



Fragments of lung tissue, from a case of phthisis of about a year's duration. *a*, mucus corpuscles and granular matter. p. 279.

Fig. 2.



Fragments of air cells, with elastic tissue from sputum. From a case of phthisis, in which the physical signs were not at all marked. × 215. p. 279.

Fig. 3.

Healthy bronchial sputum: mucus corpuscles, epithelium, and oil globules. × 215. p. 275.

Fig. 4.

Fragments of pulmonary tissue, from sputum. × 215. p 279

Fig. 5.

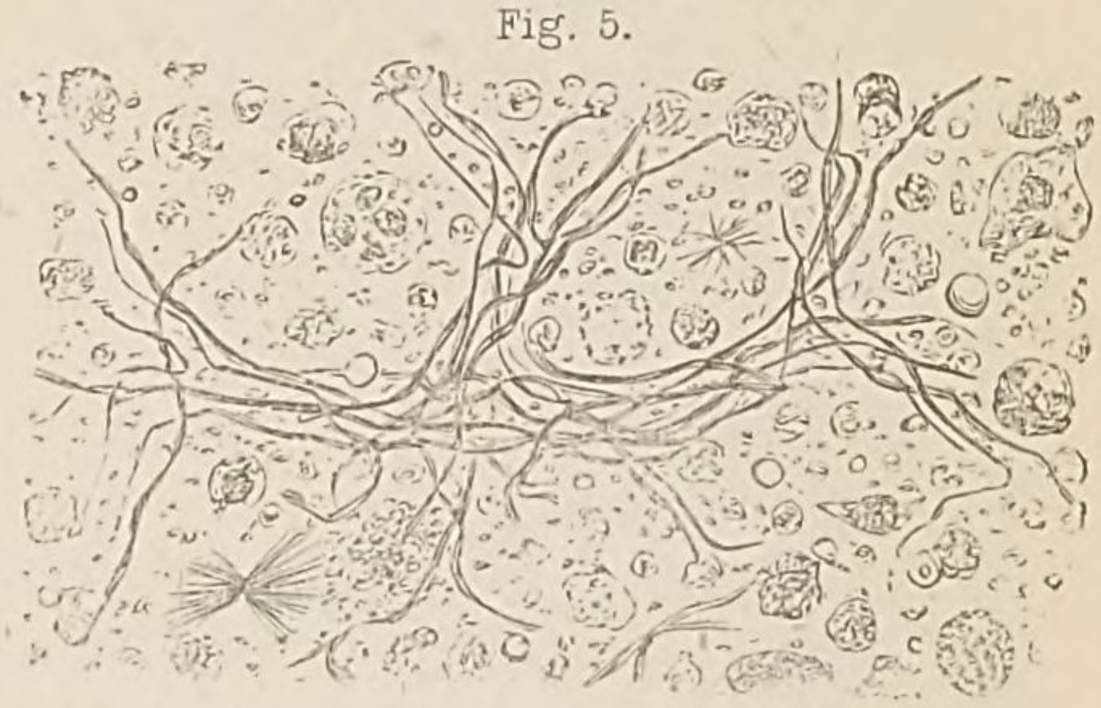
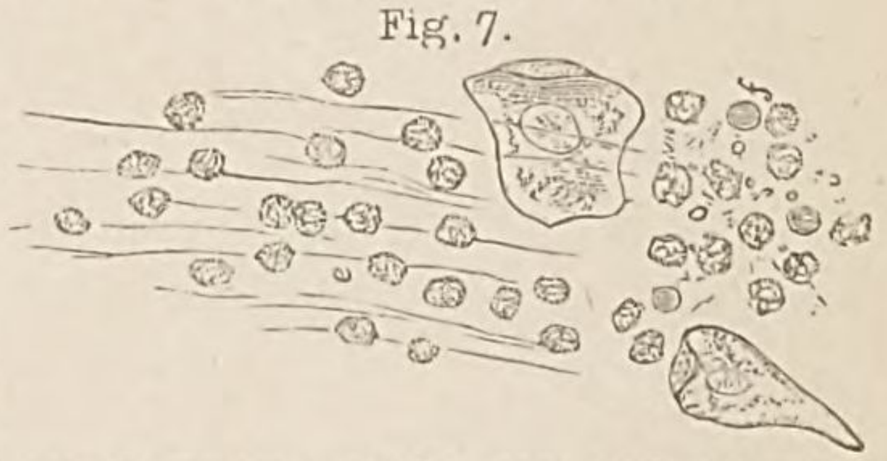


Fig. 6.

False membrane from a case of diphtheria on the fourth day. × 215. p. 285.

Fig. 7.

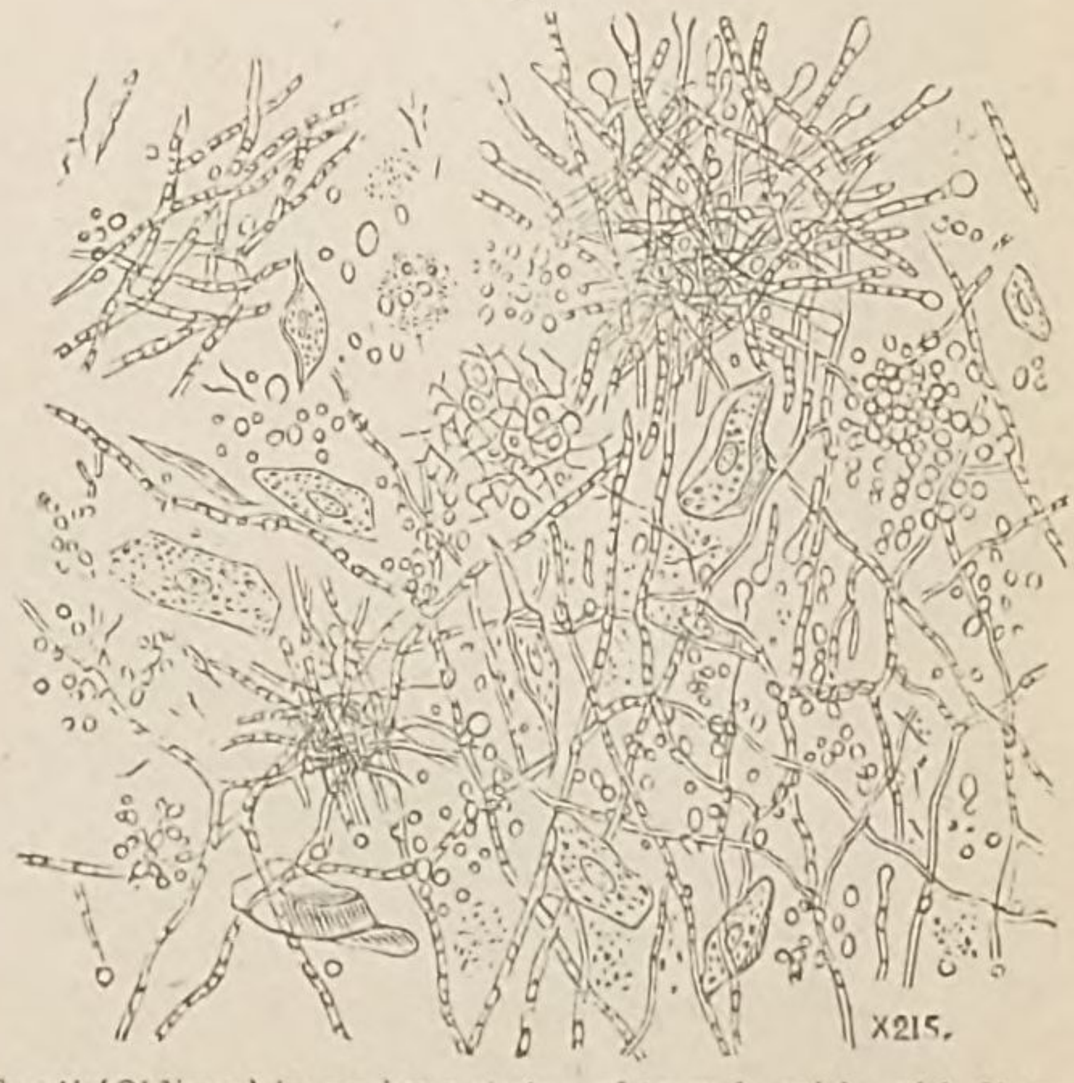


False membrane from another case of diphtheria, on the fourth day. × 215. p. 285.

Fig. 8.

Myelin present in large quantity in the sputum of a case of slight hæmoptysis. × 215 p 282.

Fig. 9.



Fungi (Oidium) in various stages of growth, with epithelium of the mouth, expectorated by a patient in the last stage of phthisis. p. 281.

1000th of an inch ,___, × 130.

" " ,____, × 215.

[To face page 284

membrane, until a firm thick lamina results, which may be detached in flakes from time to time, and which may be separated from the epithelial surface upon which it lies..

The description given above, results from observations made by myself upon specimens which have fallen under my notice. The two cases from which the drawings represented in figs. 6, 7, pl, XL, were made, occurred in the practice of Mr. Woody, of Tamworth, whose former assistant, Dr. Spratly, I have to thank for the specimens and careful notes of the cases.

Fig. 6 was obtained from the fauces of a gentleman about forty, on the fourth day of the disease. *a.* Epithelial cells from the mucous membrane of the mouth. *b.* Portion of false membrane exhibiting a striated appearance and entangling numerous cells resembling pus corpuscles. *c.* Cells like pus corpuscles, showing nuclei very distinct. *d.* Another part of the false membrane stretched somewhat and entangling corpuscles rendered oval by the pressure.

Fig. 7 was obtained from a case on the fourth day. *e.* Granular cells more disintegrated than those represented at *c*, and not exhibiting nuclei. *f.* Blood corpuscles. *g.* A portion of the mass entangling granular cells acted upon by acetic acid.

Virchow showed that exudation occurred into the substance of the mucous membrane itself, and that the tension so caused at length led to ulceration. There can be no doubt that this is so in some examples of the disease. That the mucous membrane itself is affected as well as the epithelial surface is rendered probable by the loss of sensibility in the nerve-fibres, but it must not be supposed that the "false membrane" is altered mucous membrane, for in none of the many fragments of false membrane which I have examined have I found any structures such as capillaries nerve-fibres and areolar tissue which enter into the formation of mucous membrane.

The "exudation" is not unfrequently poured out principally from the orifices of the glands and caused to spread over the epithelial surface. In the case of a very thick and firm false membrane formed upon the surface of the tongue and pharynx of a diphtheritic pig, I found that the adventitious tissue which was a quarter of an inch in thickness could be raised somewhat without being torn. In this way, funnel-shaped processes, as much as a quarter of an inch in length, were traced from the deep surface into, and to the very bottom of, the glands of the mucous membrane.

In some cases of diphtheria a great part of the epithelial layer, modified by altered growth, and by being interpenetrated by matters exuded from the blood, is stripped off in a membranous form. The epithelial layer is increased in thickness by the rapid development of new cells having the characters above described upon the under surface of the

mucous membrane. These new cells, corresponding to the deepest layer of epithelium, lose more and more the epithelial character, and tend gradually to pass into pus corpuscles. This morbid change is, however, not characteristic of diphtheria, and this disease may pass through all its stages, and end fatally without the detachment of any epithelium at all.

In many cases sporules of fungi are met with, but many circumstances tend to prove that they merely grow in the false membrane as in a nidus favourable to their development, and are not to be regarded as the cause of its production. Of course the changes already referred to involve alterations in the organic fluid stagnant in the connective tissue, amongst the epithelium, and in the capillaries and lymphatics of the part. These alterations favour the growth and multiplicaton of minute living organisms which have been called microzymes and micrococci, and which are related to the omnipresent bacteria. To the influence of such living organisms it is now the fashion to attribute diphtheria, as well as many other diseases. It is curious to observe how very easily in these days an untenable doctrine may be forced into notoriety, and taught far and wide as if it were actually demonstrated truth. A few authorities perhaps in Germany graphically portray what they please to call the results of observations, and after marshalling before the reader certain facts and arguments, remark that the evidence is perfectly conclusive in favour, say, of the view that certain contagious diseases are due to microzymes. The discovery is taken up by the "press." Papers, with "new observations," soon follow, and confirm the original statements in every particular. Pupils, friends, admirers, accept and diffuse the new doctrine. Abstracts and memoirs multiply, and the conclusions arrived at abroad are supported and promulgated here, under the patronage of a Government official, and published in a blue book. Those unacquainted with the art and mystery of transforming arbitrary assertions into scientific conclusions are easily convinced that the whole scientific world is agreed upon this one question at any rate, while in point of fact the speculative and far-fetched arguments would not have withstood careful and intelligent examination. The "facts" were not facts, and many of the "observations" were but conjectures.

The evidence recently advanced in favour of the doctrine that diphtheria is due to bacteria, or to some form of vegetable organism, is inconclusive, and rests on no solid foundation. The doctrine itself would have stood little chance of being accepted had not the scientific mind been undergoing careful preparation for years past, in order to fit it for the reception of that most extraordinary of modern scientific revelations, the bacterium disease causation hypothesis.*

Epithelial Tissue detached in Layers.

277. Detachment of Flakes of Epithelium from the Mouth, Tongue, and Œsophagus.—The practitioner is sometimes surprised to find among matters said to have been expectorated membranous shreds of firm consistence. Upon general examination he may mistake such bodies for some altered constituents of food, but by the use of the microscope he will prove that the entire mass is made up of epithelium. I have seen many examples of these curious epithelial flakes which had been detached from the tongue, fauces, and gullet. They consist, however, of the superficial layers only, and may be well compared with the large flakes of cuticle which are commonly detached during the "peeling" which occurs in three or four weeks after an attack of scarlet fever.

The epithelium is usually fully formed, and sometimes there is evidence showing that it has been produced in undue proportion, for the epithelium forms a layer, at least twice as thick as it ought to be. Indeed, I have seen laminæ peeled off which were thicker than the entire epithelial layer would be in health. In all these cases a considerable layer of epithelial tissue invariably remains behind attached to the corium. If this were not so, the peeling off of these flakes would inevitably result in the destruction of the mucous membrane corresponding to the detached portion of epithelium.

Flakes of Epithelium from the Tongue.—A man was some years ago under my care who was suffering from the formation of what appeared to be a very thick false membrane on the side of the tongue. It was moderately adherent and proved to be in continuity with the deep layers of epithelium. It came off within one or two days of the man's admission into the hospital, leaving one or two superficial ulcers. It consisted principally of altered epithelium with granular cells, sporules of fungi, and débris (Case Book, vol. i, p. 218).

Flakes consisting of a thick layer of the Epithelium of the Œsophagus.—Portions of membrane are sometimes detached from the lining membrane of the gullet and rejected. Mr. Wood, of Shrewsbury, sent me a very remarkable specimen of this some years ago. It was in the form of a distinct membranous tube several inches in length.

* It is not necessary to review the observations of Billroth, Hueter, E. Verson, Nassiloff, Trendelenburg, Letzerich, Oertel, Eberth, and Dolschenkow. Those interested will find an abstract of the papers of these authors by Dr. Sanderson in Mr. Simon's "Report to the Privy Council, 1874." After devoting nearly seven closely printed pages to the consideration of the views of the above authorities, it is surprising that the only conclusion arrived at should be that "the subject is still in some obscurity."

The following is an extract from Mr. Wood's note :—"I feel induced to trouble you with the enclosed, although I know so little about it. It was given to me by a medical friend in Shrewsbury. All I know is, that a lady patient of his was for a long time troubled with sickness which nothing would allay, and was reduced to extremities, when she vomited several pieces like the enclosed, but much longer, three inches in length. They appear membranous, but they are not, I believe, of a vegetable nature. On being burned, they give out the peculiar smell of animal tissue. I have never seen anything like this; the thicker portion feels like the membrane of diphtherite, but the thin, firm membrane is very peculiar. Is it an exudation from the œsophagus? I have just received another portion seven inches long which appears to be a cast of the œsophagus, and when fresh was of a light skin colour."

Upon microscopical examination, it was found that Mr. Wood's conclusion as to the nature of these membranous masses was quite correct, they were composed of the firm and matted layers of squamous epithelium which constitute the lining of the œsophagus. Several of the masses formed complete tubes. The case is especially interesting as demonstrating the fact that a considerable thickness of an epithelial layer may be removed from a surface which in the healthy state is only covered with a little mucus, and whose epithelium grows very slowly and undergoes little change. Cases are referred to further on in which large flakes were removed from the mucous membrane of the stomach, small intestines, uterus and vagina, and examples of detached laminæ of epithelial tissue from other mucous membranes will be found in the proper place. *See* page 297.

Examination of Vomited Matter.

278. Examination of Vomit.—As vomit usually contains a vast number of substances often more or less isolated from one another, it becomes necessary to examine several specimens taken from different parts, in order to ascertain the general microscopic characters of the whole mass. Portions may be removed upon the point of a knife; by a pipette with a very wide opening. if the vomit be not very viscid; and with the aid of scissors and forceps, if it be very thick and ropy, as in the case of sputum.

It is desirable to examine vomited matters as soon as possible after their rejection, as many substances present undergo rapid alteration. Many of the remarks, made under the head of "Extraneous Matters in urine," Ch, XVI, are applicable to vomit; and unless the observer makes himself familiar with the appearance of different substances found in food, and those which are liable to obtain entrance accidentally, he is liable to make the most ludicrous mistakes in describing the object or drawing taken from it. A fragment of feather has been described as a

lymphatic vessel, a portion of hair as a nerve tube, and other mistakes of the same kind have been made, in consequence of the observer not being acquainted with the characters of objects which he is liable to meet with in the course of such enquiries.

Vomit may be allowed to stand in a conical glass, and the deposit removed with the pipette. It may be placed on a glass slide, or in a thin glass cell, and covered with thin glass, or examined in the animalcule cage, p. 152, which is a most convenient little instrument for examining the deposits from fluids. It should be examined under a power of 200 diameters, but in this and other cases it is necessary to examine the specimen under a lower power in the first instance.

Vomit always contains fragments of vegetable and animal tissues, which have been taken as food, more or less altered by the processes of digestion. Starch globules are usually met with in great numbers; but if sufficient time has been allowed for the change to take place, the insoluble portions of the starch granules will alone remain. Pl. XLI, fig 1.

Considerable attention has been given to the appearance presented by the uredo of wheat, as it occurs in vomit, and also in stools. In the time of the cholera, the undigested uredo found in the stools was looked upon as a fungus connected with the cause of this affection, but its true nature was pointed out by Mr. Busk.

Torulæ are very frequently present in considerable numbers in vomited matters; several other forms of vegetable fungi are not unfrequently met with, and germs of bacteria, and bacteria varying in size are often very abundant. The characters of the two kinds of sarcina met with in vomit, will be described in the last chapter. See also pl. XLI, fig. 3, *g*, *h*. The vomit which contains this vegetable organism usually ferments in a very remarkable manner for some time after its rejection, like yeast, but the sarcina is occasionally found in vomit which does not possess a yeast-like appearance. Besides the sarcina, numerous oval fungi are usually present.

The colour of the so-called "coffee-ground vomit" appears to be due to the presence of a dark-brown pigment in considerable quantity, forming small aggregations or minute granules which, probably, consist of the altered colouring matter of the blood. Often a considerable number of blood globules, somewhat changed in form, are present. In some specimens of cholera vomit, numerous flocculi, consisting partly of large cells of scaly epithelium, and partly of cylindrical epithelium from the intestines, have been found.

The clear fluid which is brought up in certain cases (Pyrosis or Waterbrash) contains only a little epithelium, and a few small oil globules.

The green vomit, depending upon the presence of bile, contains cylindrical epithelium from the gall-ducts, scaly epithelium, flakes and

small masses of biliary colouring matter, often of a very bright colour, and fat globules. The crystalline fatty matter present in vomit is due to changes taking place in the fat, perhaps in consequence of the reaction of the acid of the gastric juice, pl. XLI, fig. 1. Dr. Leared attributes this change in certain cases to the action of the pancreatic fluid which in obstinate vomiting must, like the bile, reach the stomach. —Med. Times and Gazette. 1854.

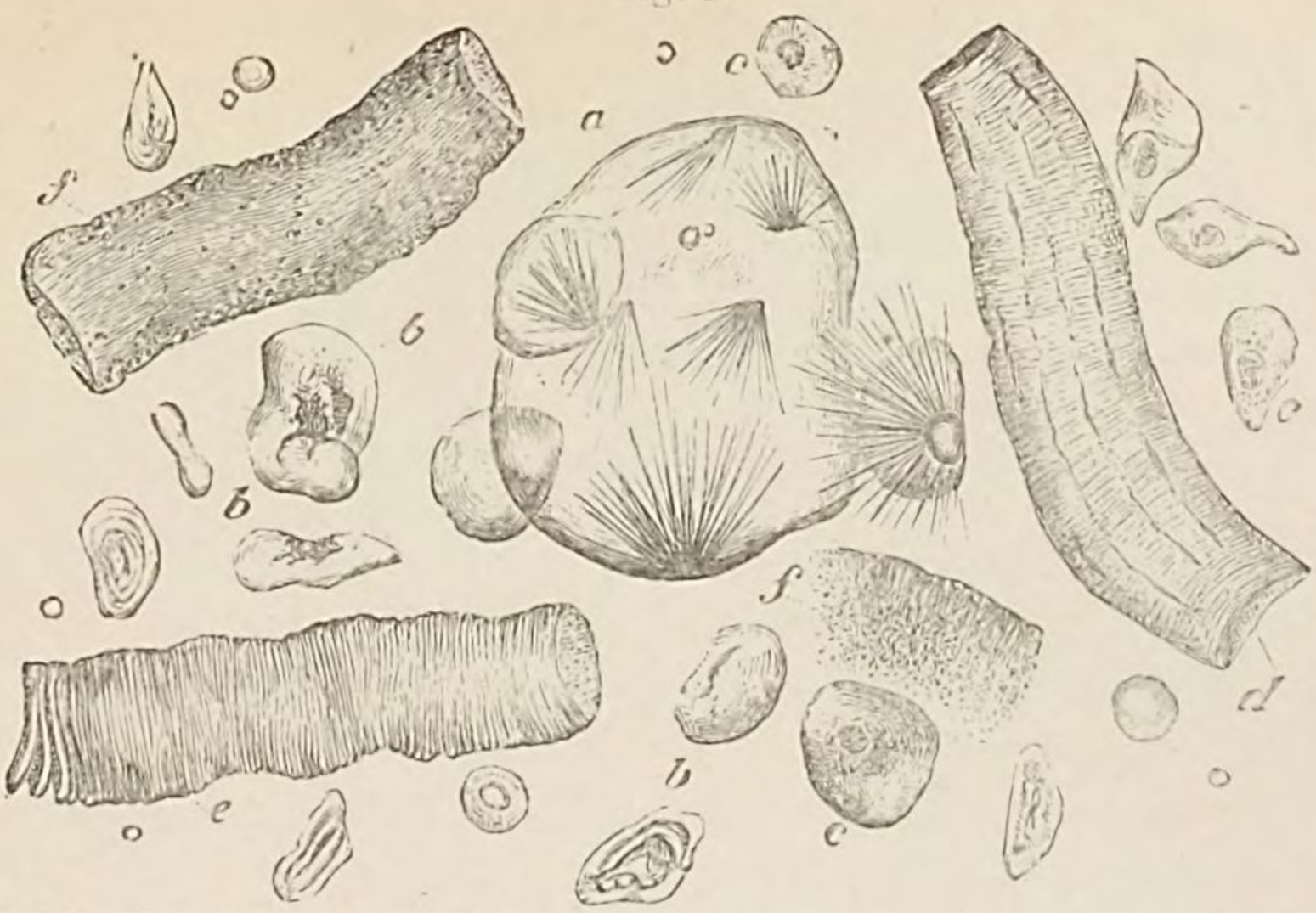
In cases in which cancer of the stomach is suspected, the vomit should always be carefully examined for cancer cells, pl. XLI, fig. 2, although usually these will be so much broken down as not to be recognizable. The observer must be careful not to mistake cells of columnar epithelium for cancer cells. It is, however, in soft cancer that cells are most commonly found in the vomited matters. It is not possible to mistake these for the ordinary epithelium of the part. Sometimes portions of the growth itself are detached when the loops of vessels covered with cells only too clearly establish the diagnosis.

Flakes consisting of Epithelium of the Stomach.—On page 287 I have alluded to the detachment of flakes of epithelium from the mucous membrane of the mouth, tongue, and œsophagus. What is still more remarkable is that occasionally flakes of stomach epithelium have been rejected in vomit. A remarkable instance presented itself in a severe case of scarlet fever under my care, in which a thin membranous mass about 3 inches by 2 was rejected. It was found to consist entirely of epithelium. After the patient's death, the part of the surface of the mucous membrane from which it was detached, was discovered, and this, with the epithelial masses, were preserved permanently.

Biliary Matter.

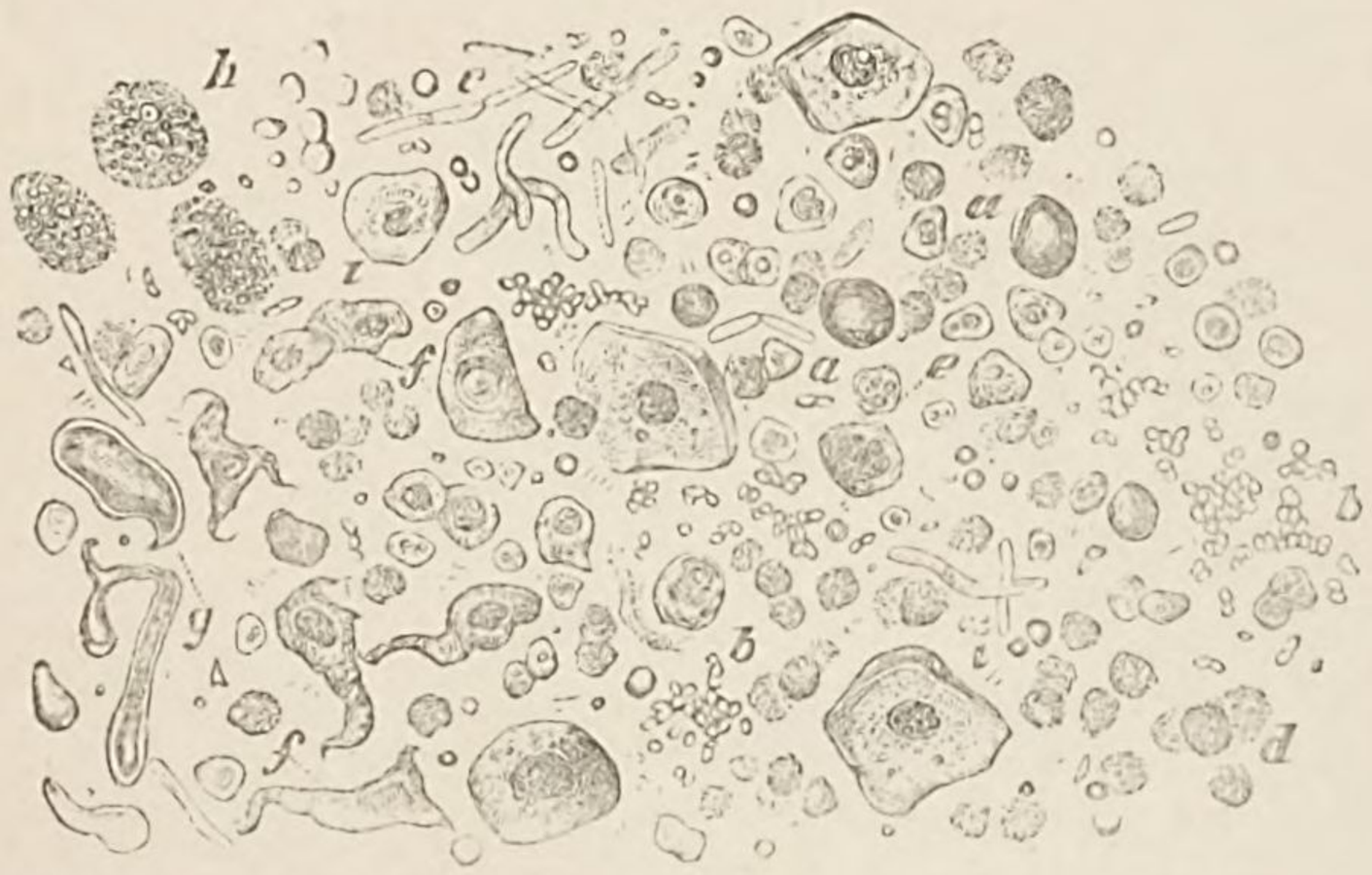
279. Examination of Bile.—Biliary matters are frequently found in vomit, and occasionally small gall stones are driven back into the stomach and rejected by vomiting. The only insoluble substances met with in bile are epithelial cells, from the ducts, of a columnar form, occasionally crystals of cholesterine, and very frequently minute dark yellow particles consisting of inspissated bile. Sometimes these are nearly spherical, almost like very minute calculi; or they may appear as little branched masses, in fact, casts of the ducts. The observer must remember that in examining the bile of many of the lower animals, especially the sheep, he may meet with the ova of entozoa, which sometimes pass into the bile in immense numbers. In the bile of fishes these are often very numerous; some of them have a very peculiar appearance, and have been mistaken for cells. Little solid particles and masses of epithelium often become the nuclei of gall stones. The mode of crystallizing bile is described in § 231, p. 195, and the acids and salts of bile are figured in pl. XXIII, p. 196.

Fig. 1.



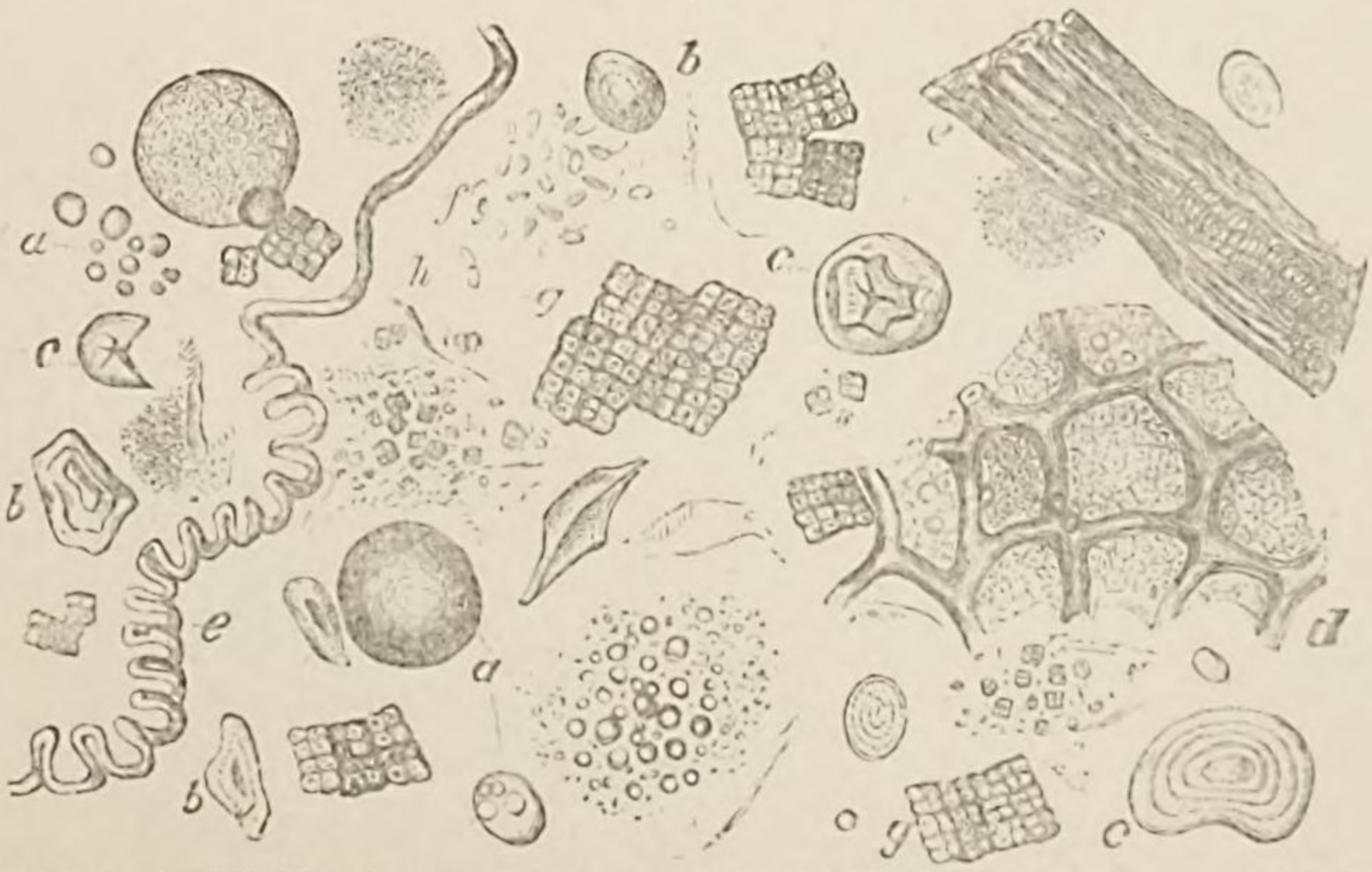
Vomit from a healthy man, three hours after a meal, consisting of bread and bacon. *a*, oil globules with crystalline fat *b*, starch granules, altered by baking and partly digested *c*, epithelial cells, probably from the pharynx. *d*, muscular fibres, scarcely altered. *e*, muscular fibre, partly digested, showing a tendency to split into discs. *f*, portion of muscular fibre, in great measure disintegrated by the process of digestion.
× 215. p. 290

Fig. 2.



Vomit from a female, age 40, suffering from symptoms of cancer of the stomach, under the care of Dr. EADE, of Norwich. The specimen was not examined until twenty-four hours after it had been rejected. The disease probably commenced upon the surface of the mucous membrane. *a*, oil globules. *b*, minute oval fungi. *c*, vibriones. *d*, pus corpuscles. *e*, cells from the surface of the ulcer. *f*, cancer cells. *g*, altered biliary matter. *h*, false cells, formed by the aggregation of numerous small particles. *i*, squamous epithelium—mouth.
× 215. p. 290.

Fig. 3.



Vomit from a patient with dilated stomach who vomited multitudes of sarcinæ. *a*, oil globules. *b*, starch granules, altered by baking and by maceration in the fluids of the mouth and stomach. *c*, starch granules (some cracked), less altered than the preceding. *d*, portion of the testa of wheat. *e*, portions of vegetable fibres. *f*, small oval fungi. *g*, sarcinæ. *h*, very small sarcinæ, growing. × 215. p. 289.

L.S.B., 1857.

$\frac{1}{1000}$th of an Inch × 215.

[To face page 290

Matters passed by the Bowels.

280. Matters passed by the Bowels.—The microscopical examination of the fæces is in certain cases of considerable importance. In dysentery, shreds of fibrinous matter, blood corpuscles, pus globules, and cylindrical as well as squamous epithelium, are sometimes present. Crystals of triple phosphate are also often met with.

Mucus casts are sometimes expelled from the large intestine, and occasionally as complete tubes. Of this an interesting example was sent me by Dr. Borrett. Flakes, some of which are very firm, are not uncommon, especially after prolonged constipation. They consist of a firm mucus, in which the epithelial cells from the large bowel, and mucus corpuscles, are embedded. The microscopical appearances were similar to those represented in pl. XLII, fig. 2.

The masses referred to above were tolerably firm, and some of them were evidently portions of a tube. They were passed by a child aged four, without giving rise to any urgent symptoms. On microscopical examination, the tissue was found to be composed of a very firm mucus, in which numerous cells of epithelium from the large intestine were embedded. The following notes of the case are extracted from Dr. Borrett's note :—"The casts of mucus and epithelium were passed by a little girl after some weeks of pain in the belly. My fears were that some foreign body had been swallowed, a button having once been passed up the nostril, and not recovered. We always had a difficulty in getting the little girl to sit down, and relief of bowels always caused more or less pain. The substance came away after a strong dose of senna tea, which caused great griping; it had been found that senna always caused distress, and castor oil was the aperient used afterwards. There was increased sensibility of the canal which made senna a bad aperient. The mucus and epithelium cast of the bowels was passed in Feb., 1860; since which time the child has enjoyed good health; she never makes any complaint of pain in the bowels; the appetite is large; and the general condition of the child excellent."

Fibrin-like Mucus.—Dr. Wynne, of Eccleshall, sent me in 1871 an interesting specimen passed by a lady who had got stout very quickly, and had suffered from hæmorrhage from the uterus. "At times this is something frightful; but if it stayed for a few hours she becomes very ill."

With regard to the passing of this substance. "It commenced two years ago for the first time, appearing and disappearing at intervals; the last time it has been so for about six weeks; but four days ago she passed more at one time than she ever did before. She can always tell when it is about to appear. Mostly the night before, when, after being an hour in bed, she begins to feel sick and very restless, and as if '*a dog was gnawing at my bones*, specially *in the wrists and ankles*,' with

a feeling of suffocation. There is great pain, but no straining at the time it is passed; and '*feels almost well again*' for a little time after it has come away."

Another case that came under my observation was a gentleman of more than seventy years of age, who also suffered from violent attacks of stomach derangement, depending upon contracting pylorus, which at length caused death. The shreds of mucus were very numerous and of firm consistence. Microscopical examination proved them to consist of hardened mucus from the large bowel. Pl. XLII, fig. 2.

In these cases the "mucus" formed is firm enough to form a tough leathery membrane. I have examined many specimens which presented the same general characters. Mucus shreds with lines, and free mucus corpuscles, and imperfectly-formed epithelial particles entangled, formed the bulk of the mass. Of course, there were bacteria and "microzymes," and plenty of minute particles which might be fungus sporules or germs; but these had nothing whatever to do with the formation of the remarkable structure referred to.

In the stools of *typhoid fever*, crystals of triple phosphate are frequently present in great number; altered blood, and vast numbers of bacteria, with different kinds of vegetable fungi, are not uncommonly found. To some of these vegetable organisms the fever has been attributed, but so far the evidence in favour of this view is imperfect and in some respects contradictory.

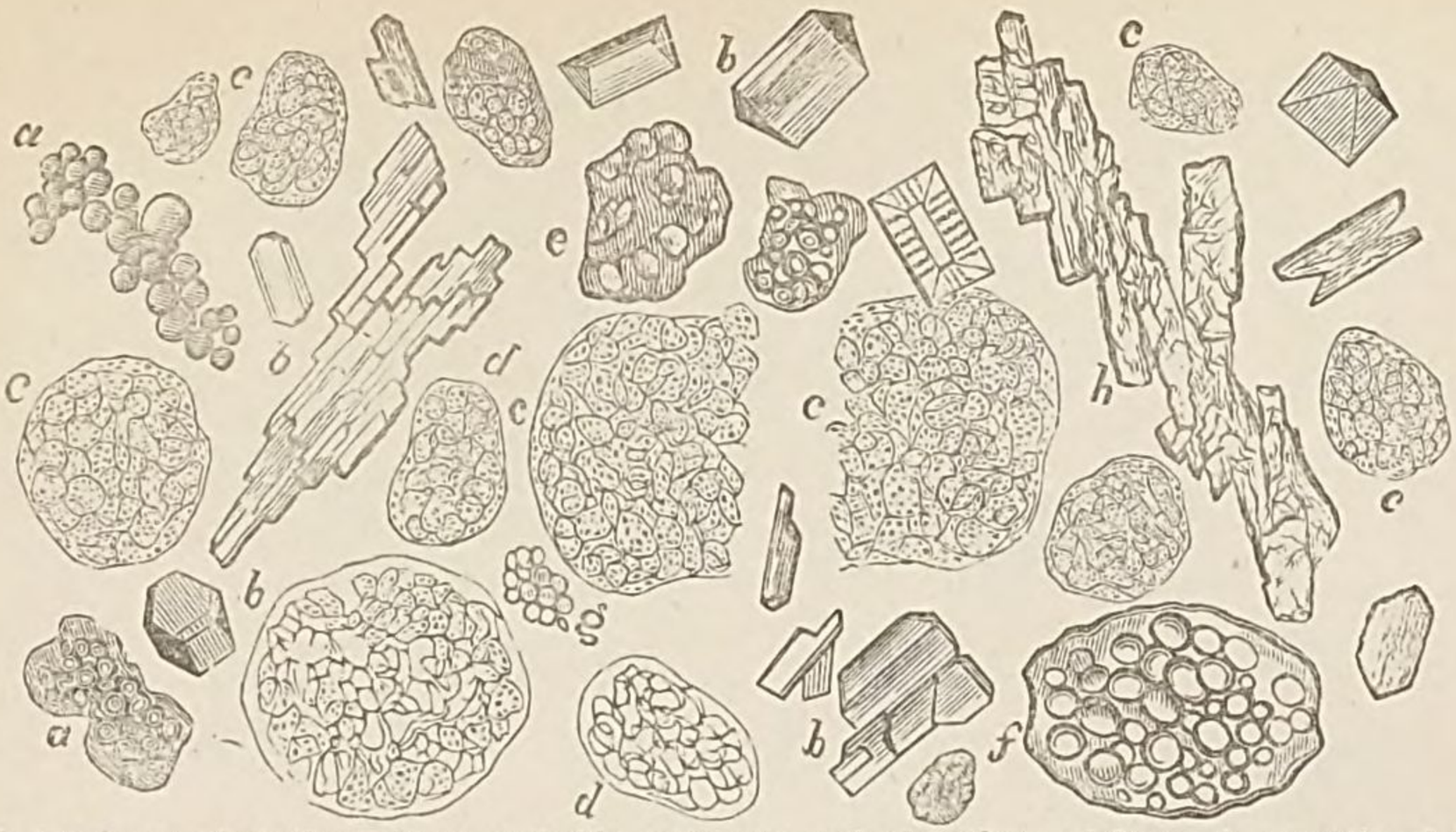
The bodies represented in pl. XLII, fig. 1, were obtained from the liquid stools of a girl aged eighteen, who was suffering from cough and fever. The oval masses are probably fragments of a clot of blood. The specimen was sent to me in 1858 by Dr. R. E. Thompson.*

The stools of cholera patients are sometimes remarkable for the large quantity of cylindrical epithelium they contain. In many instances the white flocculi are almost entirely composed of it. Sheaths of the villi are often found in great numbers. Some observers have failed to discover these sheaths, but I met with them myself in one of the first cases I ever saw, and I have seen them several times. In the majority of cases, however, according to the testimony of many observers, sheaths of the villi are not present in the stools, although multitudes may be found in the small and large intestine after death. Undigested muscular fibre exhibiting the transverse striæ very beautifully, large crystals of triple phosphate and fragments of substances taken as food, are also generally met with.

Masses of vegetable confervoid growths have occasionally been passed by the bowels, but such cases are not common; one is mentioned by Dr. Arthur Farre, and another by Professor Bennett.

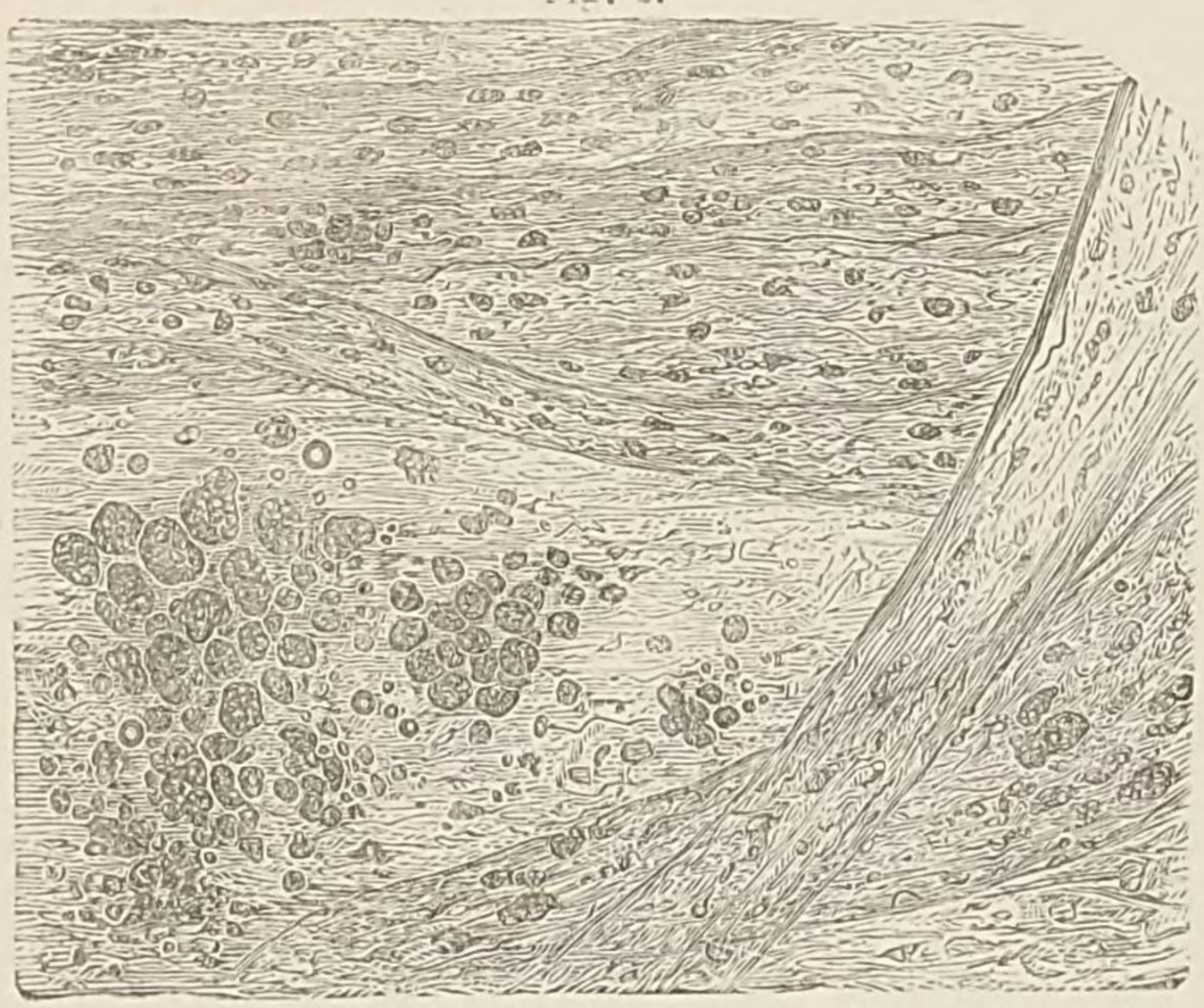
* "Archives of Medicine," No. II, page 141.

Fig. 1.



a, rounded masses of earthy matter, probably carbonate and phosphate of lime. *b*, crystals of triple or ammoniaco-magnesian phosphate. *c*, oval masses, probably fragments of a clot. In one, to the left of the figure, the outline of the blood corpuscles is more distinct than in most, and in *d* the individual corpuscles can be seen. *e*, dark amorphous masses, probably derived from the food. *f*, ovum of an entozoon, probably an ascaris. *g*, small collection of blood corpuscles. × 215 p. 292.

Fig. 2.



Mucus passed from the bowel of a man aged 70 who suffered from very chronic affection of the colon. He passed, once a month or oftener, large flakes and long tubes of tough mucus, the microscopical characters of which are represented in the drawing. The corpuscles are all modified mucus-corpuscles from the large bowel.
× 215. p 292.

Fig. 3.

Portions of the coats of a large artery which had passed through the bowels without being digested or otherwise changed Natural size.

Fig. 4.

Yellow elastic fibres, probably from an artery Found in the stools, and exhibiting very distinctly transverse markings. × 215.

$\frac{1}{1500}$th of an Inch × 215.

Professor Quekett and Mr. Brooke have met with some elastic fibres in the fæces, exhibiting the transverse striæ, which are normal in the fibres of the ligamentum nuchæ of the giraffe. The transverse division depended probably upon incipient decomposition, pl. XLII, fig. 4. The division is sometimes so distinct and complete as to have led to these fibres being mistaken for confervoid growths.

It is often desirable to determine whether biliary matters are present in the fæces. It does not necessarily follow that if the stools be pale all traces of bile acids and their products are absent. The test for bile acids is given in page 189.

281. Larvæ of Flies and other living things in the Stools.—I have found upon one occasion numerous living acari in matters passed by the bowels. I did not determine the species, nor could I ascertain positively whether they had passed alive from the bowel or had merely fallen into the dejection afterwards. Cheese mites might possibly pass through the alimentary canal without being destroyed.

I have found different kinds of maggots, the larvæ of various blow flies, in the stools, and the evidence that these will pass through the whole length of the alimentary canal in a living state is quite conclusive. See cases by Dr. Brinton and Mr. Blood, in the "Archives of Medicine," vol. iii, page 133. The beautiful drawing in plate XLIII, fig. 1, represents a larva or maggot of a fly magnified ten diameters, and in fig. 2, the larva is shown of the natural size. Such larvæ pass in a living state through the alimentary canal. Insects, or fragments of insects, can always be distinguished from other things by the presence of the air tubes or tracheæ, which will be seen if any of the tissues be carefully examined under powers magnifying 200 diameters and upwards, fig. 3, pl. XLIII. By maceration in fluids the air will be gradually expelled from the tracheæ, which are then by no means prominent objects, and might easily be passed over. The observer should therefore carefully study the tissue of insects prepared in various ways—perfectly fresh and after maceration in water, urine, and other fluids.

Milk.

282. Examination of Milk.—The ordinary microscopic examination of milk is not attended with any difficulty. All that is necessary is to place a drop upon a glass slide, and cover it with a piece of thin covering glass, employing slight pressure so as to get a very thin stratum. The general characters of the oil globules, and the fact of their not running together, and forming larger globules when pressed, should be noticed, pl. XLIII, fig. 4. Coalescence is prevented, it has been said, by the casein investment which surrounds each globule. That each globule has an investment may be demonstrated as follows. If the drop of milk be treated with a little acetic acid, the form of the globule

is much altered, and if the acid be strong, the thin covering will be dissolved, when several of the globules will run together, forming a large globule. Again, it will be found that ether will not dissolve the oil globules of the milk unless a little carbonate of soda, or some other alkaline salt, or an alkali, capable of dissolving this membrane, be previously added, when the ether immediately effects the solution of the oily matter. This instructive experiment may be performed in a test tube, or upon a glass slide, under the microscope; the reagents being most conveniently applied by using the little bulbs, page 167.

The colostrum, or the milk which is secreted first after delivery, will be found to contain many large and nearly spherical bodies, consisting of a collection of oil globules embedded in soft casein matter, resembling those which are floating free in the surrounding fluid.

By microscopical examination, the most common adulterations of milk can be readily detected,—such, for instance, as chalk or flour (starch globules). It has been said that miik has been adulterated by the addition of sheep's or other animals' brains. Such cases I have never met with, and I should think could only have occurred once or twice, as brains could not be mixed to make a fluid, either in appearance or taste, like milk. It is more probable that the adulteration in question is altogether hypothetical. If present, however, brain matter would be at once recognised by the particles of myelin, which could not be mistaken for anything else. See pl. XLIII, fig. 6. Fragments of vessels, nerve-tubes, and cells, would be readily detected upon microscopical examination. The commonest adulteration of milk is water, which is added in certain proportion, to what perhaps was originally skimmed milk. This most dishonest practice no doubt exists to a deplorable extent, although now punishable by law. Occasionally patients mix milk with urine, saliva, or tears, in order to impose upon us. The oil globules with their envelopes of casein, the precipitation of casein by the addition of acetic acid, or when the fluid becomes acid, enable us to pronounce upon the nature of the fluid.

Fungi develop in milk as soon as change commences. In hot weather milk begins to "turn," or get sour in a few hours after it has been taken from the living animal—nay, cases have been recorded in which the milk was decomposed while it yet remained in the mammary gland, owing probably to the secretion being unheaithy. I have seen bacteria in human milk at the time it was removed from the mother's breast. There can be no doubt that in certain unhealthy conditions of system the milk secreted is prone to rapid change, and not unfrequently the milk begins to decompose soon after it has been swallowed by the infant. Intestinal derangement and sometimes serious illness is the consequence. The entire alimentary canal may be occupied by a mass of half-clotted milk, every drop of which is crowded with millions of

bacteria. It is scarcely necessary to state that the evidence in favour of these being produced from bacteria germs, whose rapid multiplication was due to the surrounding conditions being favourable, is conclusive. Arguments in favour of the doctrine that they have been generated from certain particles of the milk have been advanced over and over again, but these arguments, as well as the "facts" upon which they are supposed to rest, are most inconclusive. It has even been recently affirmed that the vegetable organisms which grow during the lactic acid fermentation really spring from the oil-globules of the milk. Dr. Bastian, I believe, still maintains that living organisms really originate from lifeless particles. I should not have considered it necessary to notice such a statement here had it not been recently supported by Dr. Bastian, who has been at great pains to drag it from the obscurity in which it has long reposed, and where it certainly should have been permitted to remain in peace. Dr. Bastian republishes the drawings, which clearly show the appearances familiar to every practised observer, but which have been considered by Turgot and by Dr. Bastian to prove that the mycelia of the fungi grow from and are connected with oil-globules—a conclusion which is quite unjustifiable. It ought to have occurred to both observers that the facts observed could be well accounted for upon the much more probable supposition that the fungus sprang from a fungus germ which was hidden by the dark outline of the highly refracting oil-globules. The hypothesis that a fungus, or any living thing whatever, grew from an inanimate particle of oil, is opposed to very many facts known in connection with living beings, while the conclusion mentioned is the only inference that could be drawn by any unbiassed observer who had himself carefully studied under moderately high powers of the microscope the class of organisms in question. The assertion that living fungi grow from oil-globules will not help the cause of abiogenesis, for it is certain that it is untrue. It convicts its author of carelessness, inexcusable in anyone who has mastered the elements of microscopical observation.

In pl. XLIII, fig. 5, the appearances observed in the case of a drop of milk from a cow suffering from cattle-plague, are represented. Numerous particles of free bioplasm, which in the specimen had been stained with carmine, are seen in different parts of the field. These are very sparingly found in the milk of healthy animals, and when present contain oil-globules. They are, in fact, bioplasm particles concerned in the formation of the fatty and other constituents of milk, which have accidentally escaped from the follicles of the gland at an early stage of their formation. Now, in fever, the normal changes do not take place—the bioplasm grows with unusual rapidity, and the living particles multiply. Instead of the bioplasm of each particle being slowly resolved into the formed materials characteristic of milk, the latter are not produced at all, or in a modified form. The changes

affecting the bioplasm of the cells of the lacteal glands in fevers and inflammations are of the same character as those observed in connection with the bioplasm of glandular and other textures generally.

The very minute transparent particles of bioplasm seen here and there in the figure are in all probability the actual particles of contagious bioplasm, which are the agents concerned in transmitting the disease to other animals. In many cases in which disease is produced in man and animals by milk, minute bioplasm particles similar to those above referred to may be detected. In foot and mouth disease the milk is rendered unfit for food, and such particles exist in it. Sucking pigs and calves fed upon such milk invariably died.—See "Milk in Health and Disease," by A. Hutchinson Smee (Edward Newman, Devonshire Street, Bishopsgate, 8vo., 1875), a work in which will be found many valuable observations upon the variation of the constituents of milk.

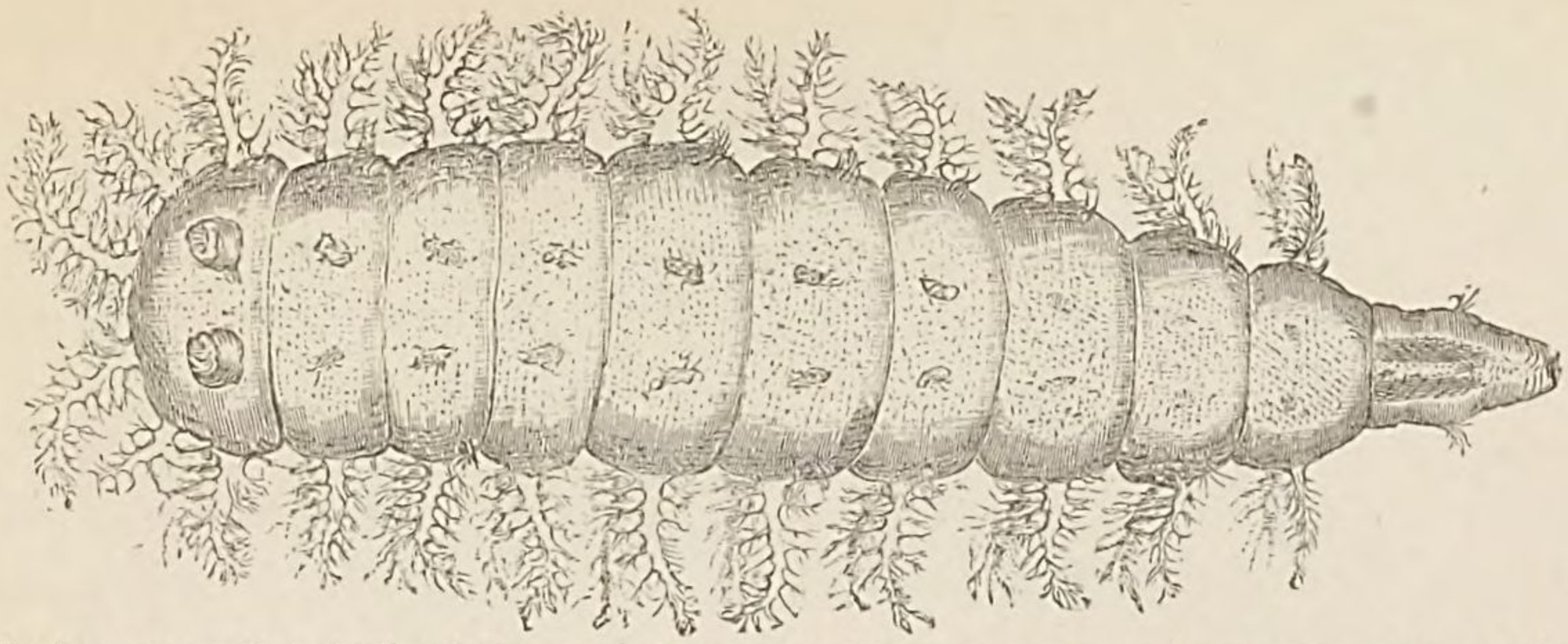
There is no doubt that milk has been the medium by which the *contagion* of typhoid fever has been introduced into the organism. In the Marylebone "epidemic," which occurred in the summer of 1873, the source of the poison was discovered. The milk supplied to the sufferers was traced to a certain farm, and from the investigations of Dr. Murchison and Dr. Whitmore there could be no question as to the way in which the typhoid germs obtained access to the milk cans. Probably no animal fluid is better adapted for the growth and multiplication of these particles of morbid bioplasm than is milk in very hot weather. At a temperature of 80°, the contagious particles would probably grow and multiply quickly. Since the occurrence of the above remarkable instance of the introduction of typhoid germs into the organism in milk, several other cases of the same kind have been recorded, and there seems to be good reason to believe that contagious fever germs have been diffused in this manner in several instances. It is, however, upon every ground of the utmost importance that whenever suspicion arises the fact should be ascertained and considered with the greatest care, and the reputed evidence very thoroughly investigated before action is taken, or very great injustice may be inflicted.

Matters from the Uterus and Vagina.

283. Discharges from the Uterus and Vagina.—The character of these discharges varies very much. In subjecting them to microscopical examination, it is better to avoid the addition of water or other fluid if possible.

In uterine and vaginal discharges, the following substances are not unfrequently met with. Epithelium of the vagina, pus globules, blood corpuscles, small transparent oval or circular granular cells, usually occurring in abundance in the mucus about the os and cervix uteri, and

Fig. 1.

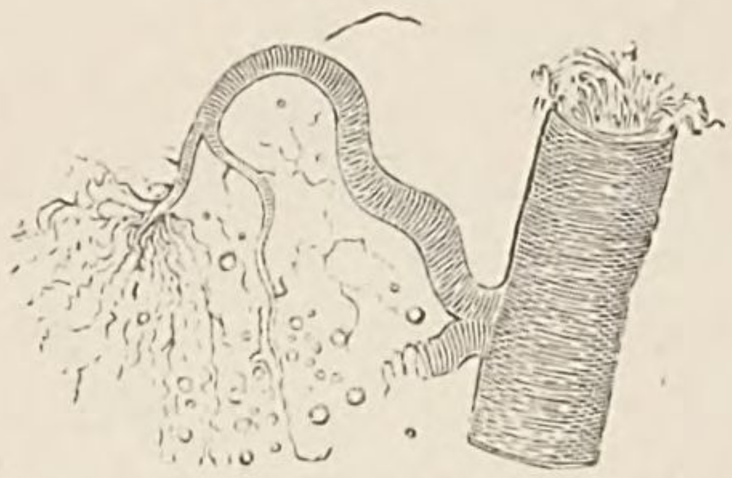


The larva or maggot of a fly which passed through the alimentary canal in a living state. × 10. p. 293.

Fig. 2.

Larvæ like the one represented in Fig. 1, but of the natural size. p. 293

Fig. 3.



Tracheæ from the blow-fly. × 215. p. 293.

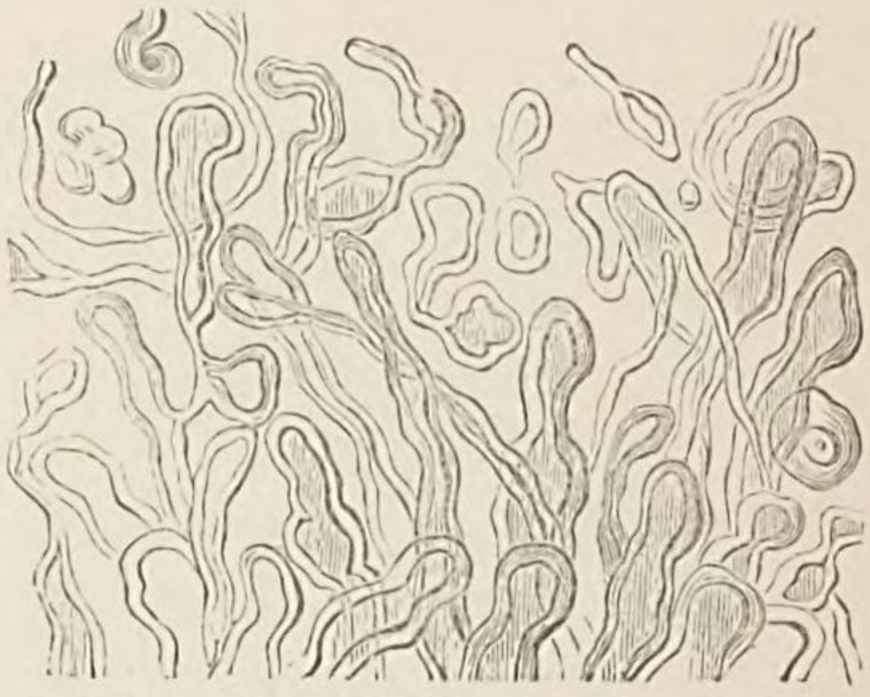
Fig. 4.

Oil globules from Milk × 215

Fig. 5

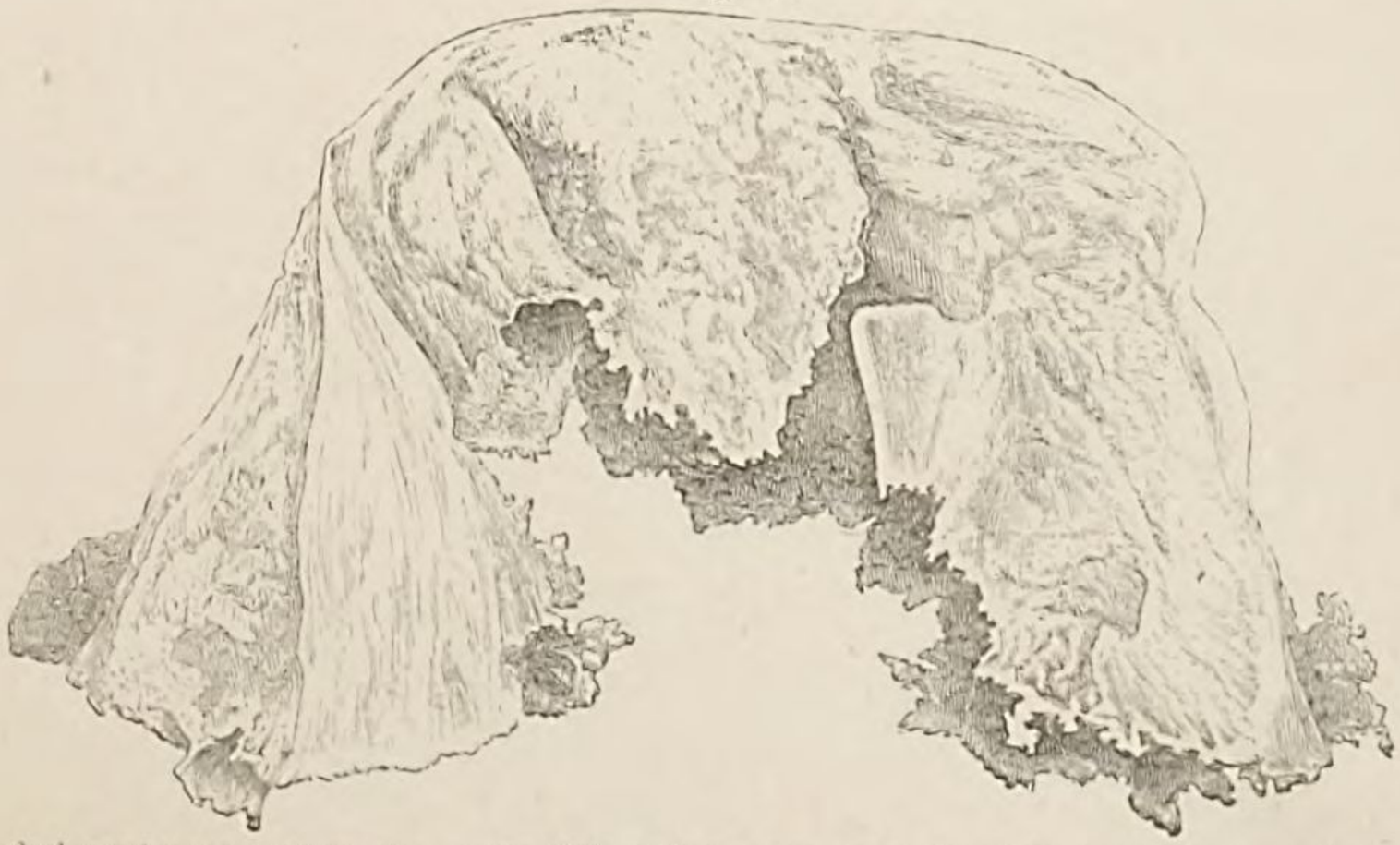
Milk. *a*, Colostrum corpuscles with bioplasm. *b*, oily matter. *c*, growing bioplasm, the so-called pus like corpuscles. From the milk of a cow suffering from cattle plague. × 700. p. 293.

Fig. 6.



Myelin from the brain. × 215. p. 291.

Fig. 7.



Epithelial cast or mould of the vagina (?), but supposed by Dr. Tilt to come from the uterus. Natural size. p. 207.

$\frac{1}{1000}$th of an Inch |—| × 215.

" " |———| × 700.

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small oil-globules. Upon the microscopical characters of leucorrhæal discharges, the Memoir of Dr. Tyler Smith, in vol. xxxv of the "Medico-Chirurgical Transactions," should be consulted.

A considerable thickness of the epithelial layer of the vagina, and according to some observers, also that of the uterus, is sometimes shed in the form of a membranous cast or mould. I have seen such epithelial casts or moulds from the rectum, œsophagus, and from the stomach. As already mentioned, they correspond to the layers of cuticle which are detached from different parts of the cutaneous surface after scarlatina.

Dr. Arthur Farre has recorded some interesting cases of "Exfoliation of the Epithelial coat of the Vagina," in vol. i of my "Archives." The appearance of the specimens is represented in pl. XII of the "Archives of Medicine." Dr. Farre remarks that the act of exfoliation is repeated at intervals. The casts described by him are interesting in another point of view, as showing the real form of the vagina when in its ordinary empty and collapsed condition. (See "Archives," vol. i, p. 71.) Dr. Tilt has also described some interesting cases of the same kind. His opinion is, that some of these cases come from the uterus, while others are formed, as Dr. Farre stated, in the vagina. The beautiful specimen figured in pl. XLIII, fig. 7, is one of those examined by Dr. Tilt, and considered by him to come from the uterus, although the characters of the epithelium of which it was composed, agree more closely with those of the vaginal cells. (See "Archives," vol. iii, p. 26.) Dr. Beigel, of Vienna, has recently published an elaborate memoir upon the same subject. "Zur Pathologie der Dysmenorrhœa membranacea." Leipzig, 1876.—A. Engelhardt.

Leuchorrhæa.—In this condition very many imperfect cells of vaginal epithelium are formed upon the surface of the mucous membrane, as well as pus corpuscles, fig. 4, pl. XLIV, page 298. The development of pus corpuscles from the bioplasm of epithelium may be studied in any case of leuchorrhæa very successfully, even after the epithelial cells have assumed their distinctive form. Many of the younger cells of vaginal epithelium, and those in the follicles of the mucous membrane, divide and subdivide, giving rise at length to multitudes of the spherical granular cells we know as "pus corpuscles," which divide and subdivide very rapidly if freely supplied with nutrient matter, figs. 4, 5, 6, 7, 8.

Cancer.—In cases of cancer of the uterus, we should expect to meet with cancer cells in the discharge, but these are often so broken down as not to be recognisable; still, when this condition is suspected, the discharge, and also the urine, should be subjected to very careful and repeated microscopical examination. I have seen marked examples of cancer in the urine, pl. XLIV, fig. 1, and occasionally I have found vascular loops detached from the morbid growths. Sometimes villous

processes, with loops of vessels, and the modified epithelial coverings, are separated entire. In this investigation, the resemblance of the cells of columnar epithelium from the ureter, to spindle-shaped cancer cells, must be borne in mind, and the student must be careful not to mistake the former for the latter. In many cases it is not difficult to remove a little of the softened cancerous matter upon the extremity of the sponge used in vaginal examinations. In this way a much better chance of meeting with entire cancer cells is afforded than by examining the urine.

It is not uncommon to detect cells containing much dark granular matter extremely like cancer cells in the urine. In some cases I have found that the dark granules consisted of particles of urate of soda, but in others they were composed of pigmentary matter. Some of these cases are extremely difficult of diagnosis. I have figured a urinary deposit from a case of acute renal inflammation, the cell elements of which might be easily mistaken for cancer-cells. See chapter XVI.

CHAPTER XV.

PUS.—ANIMAL POISONS.—TUBERCLE.—*Examination of Pus; Micro-scopical Characters.—New Observations upon the Pus-Corpuscle.—The changes occurring in Living Pus.—Of the relation of Pus to Lymph Corpuscles and Lymphatic Vessels.—Properties and Powers of Normal Bioplasm and of Pus descended from it.* OF THE POISON OR VIRUS OF CONTAGIOUS FEVERS.—*Of Contagious Bioplasm.—The Bacterium Hypothesis.—Cohn's Views.—Lister's Views.—Real Nature of the Contagious Bioplast.* OF TUBERCLE.—*Examination of Tubercle.—General Characters of the Tubercle Bioplast.—Of the "Giant Cells."—Growth and Multiplication of Tubercle Corpuscles.—Origin of a Tubercle.—Degeneration.—Caseation.—Fibroid Change.*

IN this chapter I shall consider the methods of examining three very important forms of morbid matter, *pus*, *contagium*, and *tubercle*, and I shall refer to various points bearing upon the nature, origin, and propagation of these active particles. The views I have been led to entertain have been founded upon facts and observations which will be briefly noticed, and which may be demonstrated and repeated by the student.

284. General Considerations.—The pus-corpuscle which is formed in inflammation, *the disease-germ* or contagious particle, which is the active material concerned in spreading contagious diseases, and *the tubercle corpuscles* are, as I have shown, different forms of bioplasm or living matter. All have been, as I believe, derived from, and are therefore closely related to, the living matter or bioplasm of man himself or to that of the higher animals. I shall also adduce evidence to show that neither of these bodies is a vegetable organism, and that neither has descended from any other low form of living matter outside or independent of the human organism. It seems to me that if certain important facts be considered, the conclusion that each has been derived by direct descent from the bioplasm of the body of man, or from that of one of the higher animals will be irresistible. Different kinds or forms of bioplasm having very different powers, characterise different animals, and different forms of morbid bioplasm as *pus*, *contagion*, and *tubercle* are formed in the different vertebrate classes, and even in different species belonging to the same class. Nay, we shall find ourselves compelled by the facts of the case to admit, that in one

species of animal, and certainly in the case of man himself, several different forms of pus-bioplasm, of contagious bioplasm, and of tubercle bioplasm may be developed. Some of these have such specific and particular characteristics that no doubt can be left upon an unbiassed mind that they are distinct forms of morbid bioplasm. As is well known, we have many special forms of inflammation, of fever, and of tubercular disease, each of which may be said to be caused by the growth and multiplication of corresponding special forms of morbid bioplasm.

A form of morbid bioplasm generated in one animal may grow and multiply and induce disease in an animal of a different class. A contagion, for example, developed in animals so far removed from man as are the horse, the dog, and the ox, may grow and multiply in man's body and cause a disease, which, though very different in its course and symptoms from the original malady developed in the animal, is, nevertheless, so distinct, characteristic, and peculiar that it can always be identified. One of the most striking as well as the most terrible of examples is the contagion of glanders, which develops in man a fatal fever differing in many particulars from the glanders of the horse whence the contagion was derived.

Although there are many different kinds of pus, of contagion, and of tubercle which exhibit special characteristics as regards the rate of growth, and the virulence of the morbid phenomena resulting, not one of these can be certainly identified by its microscopical characters, chemical composition, or any physical peculiarity, nor can any one with certainty by such means be distinguished from the rest. Not only so, but not one of these three forms of bioplasm can with certainty and in all cases be distinguished from healthy bioplasm. The identification of any particular form of bioplasm is to be determined by its *vital*, not by its *physical* properties, and it is, by its vital characteristics, that the power of morbid bioplasm to induce a simple or specific inflammation, a fever or a tubercular disease, depends. There are different kinds of inflammatory change, in all of which, however, "pus" is produced, but the particles of the pus bioplasm formed in each case may differ, not only as regards the size of the corpuscle and the rate of its growth, but in respect of its power of resisting external conditions with reference to the products resulting from its death.

There is then no test by which the bioplasm of pus can be with certainty distinguished from that of tubercle, or either of these from contagion. But no one who had thought over the matter carefully would conceive it likely or probable that any such test would ever be discovered, for if the bioplasm of living forms so distinct from one another, as, for example, that of man, the ox, and the dog, cannot be determined by any known characters or tests, how can we expect to

discover tests by which the pus can be distinguished from the white blood corpuscle, or the tubercle corpuscle from either?

As I have said, the three different forms of morbid bioplasm referred to which belong to man, originate under very different circumstances. The conditions requisite for the development of the pus-corpuscle are very different from those attending the origin of a particle of contagion of a specific fever, and both are different from the conditions which determine the origin of tubercle bioplasm. Different periods of time are required for the production of each morbid bioplasm. Pus may be developed in man and in most warm-blooded animals from almost any kind of normal bioplasm in the body, and in some cases in the course of four and twenty hours. Bioplasm having the remarkable properties of the contagious disease-germ, at least in one or two cases, may be produced in the course of a few days, but for the development of a tubercle-corpuscle from the normal bioplasm of the body, there is reason to conclude that a still longer period of time is required. In other words, certain conditions must exist and must be maintained for a considerable period of time to ensure the formation of a tubercle-corpuscle endowed with the wonderful powers which characterise that body. Such at least are the conclusions which have been gradually forced upon my mind by a careful consideration of facts ascertained by observation and experiment.

Pus.

285. Examination of Pus.—The microscopical examination of pus bioplasts is easily performed. A thin layer of the matter supposed to contain the pus is placed upon a glass slide, and lightly covered with thin glass. To prevent the corpuscles being crushed by pressure, a hair may be interposed between the glass slide and the cover. When pus is detected in a secretion, the practitioner must not draw a too hasty conclusion with reference to the nature of the case, for small quantities of pus may be present, at least in the urine, without the existence of any serious disease.

Pus corpuscles become smaller on being placed in saline solutions of greater specific gravity than the serum in which they float, in consequence of exosmose of part of their contents. The corpuscles of pus are destroyed by the action of caustic alkalies, and converted into a thick glairy mass, which cannot be poured from one vessel to another in drops. Upon examining the glairy mass in the microscope, no corpuscles will be discovered, but only a few granules will be observed. Ammonia, however, acts slowly upon pus corpuscles, while the white and red blood corpuscles are readily dissolved by it.

Water containing a trace of iodine in solution causes the pus globule to swell, and displays a more dense matter in the centre. The

most characteristic reaction, however, is produced by the addition of acetic acid. This reagent, if not very strong, causes the corpuscle to swell and to become perfectly clear. It may be nearly twice its previous diameter; its outline being very thin, though clear and distinct. From one to four little bodies are developed in the centre (figs. 2, 3, *a*, pl. XLIV). These have a dark outline, and are of a rather irregular form. They are highly refracting, but are not soluble in ether.

Occasionally I have found pus corpuscles in which the central contents were very dark, and slightly separated from the apparent "cell-wall," fig. 2. Upon treating these with acetic acid, the ordinary reaction ensued (*a*). It appeared as if the acid caused the central mass to contract, probably after dissolving part of its constituents. The central bodies have been termed nuclei, but they cannot be regarded as analogous to the so-called nuclei of cells generally.

286. Microscopical Characters of the Pus Globule, and of Detecting it.—I must here offer a few very brief observations upon the pus corpuscle, and the inferences to be drawn from its presence.

When a small quantity of pus from an abscess is placed between glasses, and examined with a power of about two hundred diameters, numerous granular particles, larger than a blood globule, but having a circular outline and finely tuberculated surface, are noticed. The serous fluid in which these bodies are suspended usually contains a few free fat globules, as well as granules of phosphate of lime, and acicular crystals, sometimes arranged in groups, composed of a fatty acid. Besides the above, *bacteria* and the germs of bacteria, *microzymes*, as they have been termed, are very frequently seen. The presence of these living organisms is an indication that chemical change has already commenced, or that decomposition of certain of the organic constituents of the pus corpuscle or of the serum around it has actually taken place. Now, if the spherical pus corpuscles themselves be carefully examined with a high power (700 diameters or upwards), many will be found in the very substance of which bacteria or minute particles, which, under favourable circumstances, would soon be developed into bacteria, will be distinctly discerned. In fact, the pus from the abscess consists chiefly of dead pus corpuscles and of the substances resulting from the disintegration of pus. The figures generally given of pus represent *dead*, not *living and growing pus corpuscles*, and the usual description also applies to pus corpuscles that have ceased to live.

If now a little pus be taken from a suppurating surface of skin, or better of a mucous membrane, many corpuscles will be discovered, the outline of which, instead of being spherical, is *most irregular*. From all parts of the surface will be seen little projections which are from time to time detached. In this way pus corpuscles multiply. With the aid of a high power, multitudes of smaller corpuscles will be discerned

amongst the large, fully-developed pus corpuscles. In short, the pus corpuscles of living pus vary very much in size, many being so very small and transparent as to require for their demonstration a high power and care in the management of the light.

Much trouble was taken in the earlier days of microscopical research to assign definite characters to pus corpuscles, in order that they might with certainty be distinguished from the so-called "mucus corpuscle," and other bodies which they much resemble. Such a distinction, however, cannot invariably be made, for, in the first place, bodies may be obtained which represent various stages between an epithelial cell and a pus corpuscle; secondly, cells agreeing in their microscopical characters with the pus corpuscle are not unfrequently formed upon the surface of a mucous membrane, without its functions being seriously impaired, and certainly without the occurrence of those preliminary changes which usually precede the formation of pus; and, thirdly, particles are found in the lymph, in the blood, in the lymphatic glands, in the serous fluid in the interior of certain cysts, and in many other situations, which in their size, form, and general appearance, so much resemble the corpuscles found in true pus, that it is quite impossible to assign characters by which they may be distinguished. The figures of these particles, as they appeared before and after treatment by acetic acid, often could not be distinguished from the figures of pus corpuscles, treated in a similar manner, given by the same authors. The so-called "mucus corpuscle" is nothing more than an imperfectly formed epithelial cell, which is surrounded by the transparent viscid mucus found by it. This mass of bioplasm, the mucus corpuscle, may grow and multiply very rapidly, and the particles which at last result will be true pus corpuscles. In inflammation of the bronchial mucous membrane we have, in fact, an excellent opportunity of demonstrating the transitional stages through which free bioplasm particles, or the bioplasm of epithelial cells, pass when they give origin to pus. A particular particle of bioplasm may form mucus which collects around it, and be properly regarded as a mucus corpuscle, or, instead, the formed material may become hard and condensed, in which case an epithelial cell results,—or, being freely supplied with pabulum, it may grow and multiply very quickly, in which case *pus* will be formed.

Cases occur in which the question of the presence or absence of pus is unimportant. If only a few globules exist, for example, in urine, it is a matter of no consequence. At the same time, it must not be supposed that, in all instances, the identification of pus is a matter of secondary importance. All that is aimed at in introducing these observations is to impress upon the student the importance of not stating that pus has been found in any particular locality, or in any particular fluid, merely because a few particles having many of the

characters of a pus corpuscle have been observed. To assert that the "pus had been found in the blood," or that "the casts of the uriniferous tubes contained pus," would lead to a very different inference from that which would be deduced from the remark, that "particles having all the characters of pus globules had been found in the blood," or that the "casts of the tubes contained particles resembling those of pus." The first would be true in extremely few cases; the last in a vast number that fall under the observation of every practitioner. If, however, we find a considerable number of corpuscles in the field of the microscope, of nearly uniform size, agreeing in general characters with the pus corpuscle, and which upon the addition of acetic acid give the characteristic reaction, we shall seldom be wrong in calling them pus corpuscles.

In examining the blood in cases in which the white corpuscles are enormously increased in number, there can be no difficulty in deciding, since we have weighty reasons for believing that pus corpuscles could not possibly exist in the blood under the same circumstances.

287. New Observations on the Pus Corpuscles.—The origin of the pus corpuscle has been already referred to in pp. 248–9. The researches upon which the conclusions are based have proved, I think, as I showed in the first course of lectures which I gave at the Royal College of Physicians in 1861, that a pus corpuscle is not formed by the breaking up of the tissue, and the aggregation of lifeless particles resulting therefrom. Neither is the pus corpuscle produced by the precipitation of particles from a clear exudation, and their subsequent aggregation to form masses, as Dr. Bennett, of Edinburgh, supposed. Nor, as has been more recently affirmed, is the pus corpuscle a white blood corpuscle which has changed its place. The white blood corpuscle is one thing; the pus corpuscle a very different thing.

Pus, as has been already stated, is a form of living bioplasm, and has been produced by continuous descent from the normal bioplasm of the body. Virchow concluded that pus was formed in connective tissue corpuscles, and in epithelial cells only. But there is no doubt that pus may be derived from other forms of bioplasm—probably from any bioplasm in the body—but from some bioplasm as that of epithelium or connective tissue, much more readily than from other forms. The white blood corpuscle, the minute masses of bioplasm which I have described as existing in the blood, lymph corpuscles, chyle corpuscles, the masses of bioplasm in the spleen and other ductless glands, those found in connection with the walls of capillaries, the bioplasm of nerve, muscle and other tissues of the body, may give rise to pus if placed under conditions in which they are too freely supplied with pabulum.

On the Changes occurring in ordinary living Pus.—I propose now to bring forward evidence, which seems to me conclusive, as to the

Fig. 1.

Cancer cells from the urine in a very bad case of cancer of the uterus. The deposit was very abundant. × 215. p. 297.

Fig. 2.

Pus corpuscles under the action of acetic acid. × 215. p. 302.

Fig. 3.

Pus corpuscles. *a*, some cells acted upon by acetic acid. × 215 p 305.

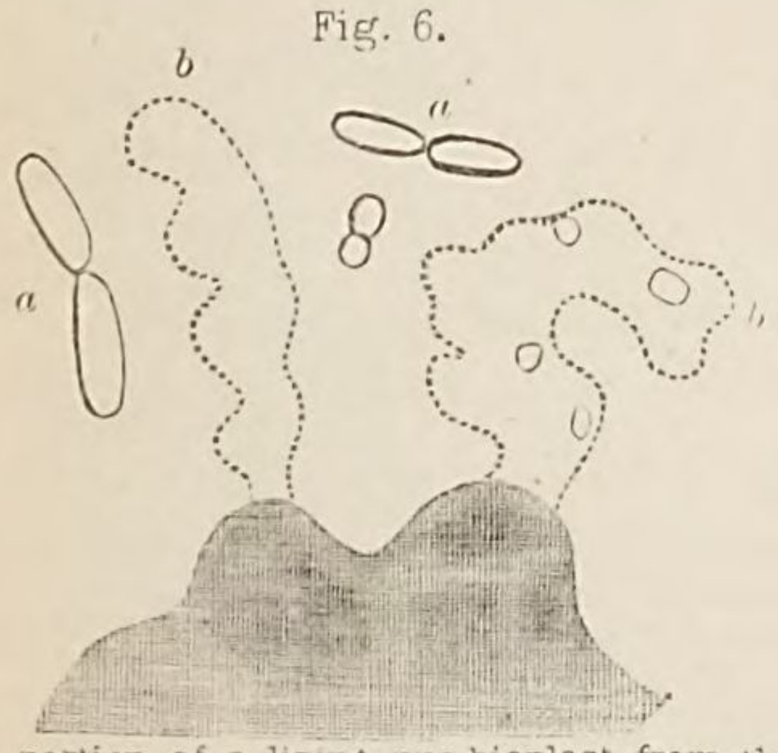
Fig. 5.

Pus corpuscles, showing protruberances. × 700. p. 305

Fig. 7.

Multiplication of pus corpuscles by detachment of protruding portions from each corpuscle. × 700, p. 305.

Fig. 6.

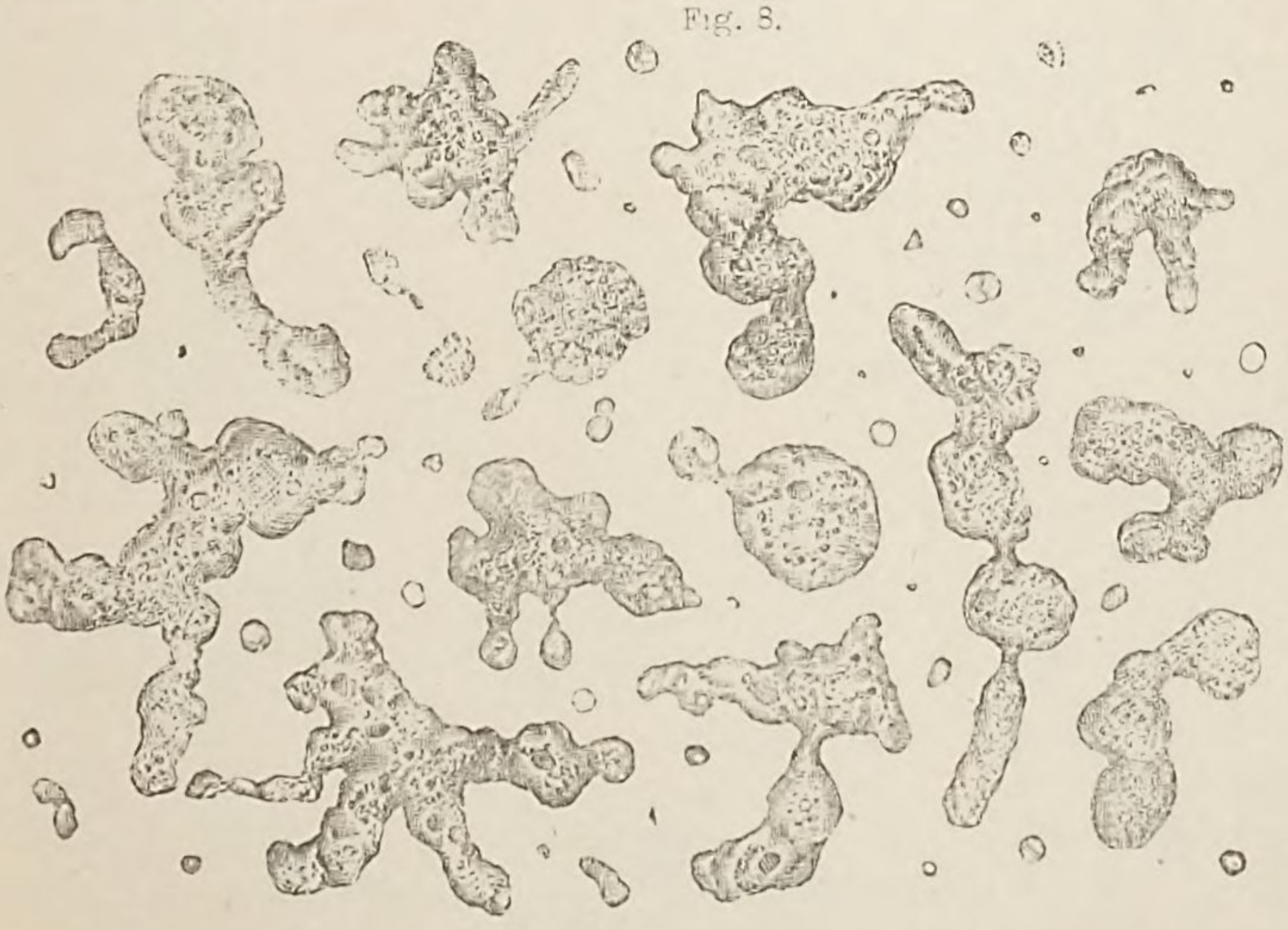


A portion of a living pus-bioplast from the bladder. The portions marked *b* moved out in four seconds, and were then retracted. *a*, Bacteria. × 2500. p. 305.

Fig. 4.

Formation of pus from bioplasm of epithelial cells of the vagina. × 215. p. 297.

Fig. 8.



Pus bioplasts in active movement, from the urine of a case of chronic inflammation of the bladder. × 700. p. 305.

1000th of an inch | | × 215.

$\frac{1}{1000}$ of an inch |——————| × 2800.

[To face page 304.

mode of growth and multiplication of pus corpuscles, and which, I think, goes far to show how living particles, so minute that they may be transferred considerable distances without loss of vitality, may be produced.

There is certainly no true cell-wall in the case of ordinary pus. This is proved by the fact that protrusions of the living matter of which pus corpuscles consist, may occur upon every part of the surface; some of these protruded portions, after moving a considerable distance away from the mass, become disconnected from the parent mass. In this way new corpuscles are produced, and rapidly increase in number. In pus from the bladder suspended in urine, containing little urea and solid matter, movements even more active than those in the mucus corpuscle can be very easily studied. As has been already remarked, in consequence of the alterations in form of the pus when fresh, *not a single spherical corpuscle can be found.* See fig. 8, pl. XLIV, representing some of the many different forms of pus corpuscles present in a very small quantity of pus. Every corpuscle exhibits a great number of these protrusions, and every protrusion might be detached and form a free pus corpuscle, figs. 5, 6. In warm weather, I have known the movements continue in pus corpuscles in urine containing little of the ordinary urinary constituents, for more than forty-eight hours after the urine had left the bladder. The very phenomena which take place upon the surface of the mucous membrane of the bladder may in fact be watched for hours under the microscope, and there are few things more beautiful or more instructive. The living matter that moves, in this and in all other cases, is perfectly transparent and structureless. Little particles which are embedded in it, and appear to move, are really moved by it.*

The formation of protrusions above referred to, and the active movements in the living matter of the pus corpuscle cease if the corpuscles themselves be moved quickly in fluid, and the corpuscles become spherical. A mass of any soft, living bioplasm when suspended in fluid in active movement, invariably assumes the spherical form, and retains it for a short time after the movement has ceased, but if placed under favourable conditions it absorbs nutriment and soon exhibits *vital movements*, grows, and multiplies. The amœba in water, the

* It is probable that careful observations upon this transparent living moving material will teach us much concerning the nature of life. I think that this subject merits far more attention than it has hitherto received, not only from physicists, chemists, and physiologists, but from philosophers. I am sure that the facts learnt from the study will by no means favour the notions now most popular, but surely that is no reason why such observations should be wholly neglected by those who profess to carry their enquiries to the utmost possible limits. Some who profess to be very liberal in science seem terribly afraid lest enquiry should be pursued a little further than happens to be favourable to their narrow materialistic dogmas.

white blood corpuscles in the liquor sanguinis, are both spherical while the surrounding fluid is in active movement, but after they have been at rest for a short time they exhibit their characteristic and very wonderful movements.

Some other changes commonly occur in the pus corpuscle. Supposing it to be subjected to influences unfavourable to this rapid growth, the material upon the surface may be precipitated, and in this way a very thin layer of semi-transparent insoluble material may be formed, which is rightly termed "*cell wall.*" Changes may then take place in the mass of the pus corpuscle itself; it may be resolved into oil globules as already mentioned, bacteria may grow and multiply in its substance, and the many alterations which are familiar to observers may be readily noticed in corpuscles which are subjected to disintegrating processes, or to the influence of fluids not fitted for nourishing them.

Many kinds have been described according to the general characters the pus exhibits, and the conditions under which it has been produced. I have before remarked in page 229, that it is not possible to distinguish many pus corpuscles from lymph corpuscles, white blood corpuscles, and many other masses of bioplasm. Nay, if the brain of an embryo be examined at an early period of development, it will be found that this important structure consists of nothing more than a number of spherical bioplasts, which could not, by any means we are yet acquainted with, be distinguished from pus corpuscles, pl. XXVI, figs. 9, 10, page 230. If we carefully reflect upon many of the observed facts referred to, we shall be compelled to admit that masses of bioplasm, which resemble one another in every character we can ascertain, differ nevertheless remarkably in *power*, as proved by the results of their living. Few recent writers appear to have fully recognised the remarkable truth that living things that agree in physical and chemical characters differ widely in *power*. The most remarkable difference in *vital power* may be associated with similarity if not with identity of composition. Physical and chemical properties neither account for the actions of living matter, nor determine the *form* of being or tissue living matter is to evolve.

288. Question of the Relation of Pus to Lymph Corpuscles, and Lymphatic Vessels.—The general resemblance which the pus corpuscle bears to the lymph and white blood corpuscle, has led many to regard these bodies as identical. That such a view should have been accepted in the early days of microscopic observation was natural; but that it should have been recently revived and confidently taught is, indeed, surprising, since every observer ought to be aware of the fact that as regards living matter resemblance in appearance is constantly associated with the most remarkable difference in properties. The fact that from any bioplasm of the body spherical masses of bioplasm, like white

blood corpuscles, may be derived appears to be almost unknown, or the doctrine that every spherical mass of bioplasm seen in a tissue is either a white blood corpuscle or a lymph corpuscle would not have been so hastily and so generally accepted. The most extraordinary misconceptions seem to prevail as to the nature and origin of such bodies, and the most extravagant conclusions have been advanced, all apparently based upon the erroneous assumption that a spherical mass of bioplasm must have come from a lymphatic, or from a blood vessel, and that general resemblance in size and form involve agreement as regards nature and origin.

More or less rounded or spherical masses of bioplasm have been looked upon as lymph corpuscles simply because they are like the bioplasm particles suspended in lymph. But as I have shown in this work and elsewhere, there is not a form of bioplasm from which such spherical particles might not under certain circumstances spring, and I do not believe a greater error can be committed than to conclude that, because certain particles of bioplasm are "like" other particles that both must have been derived from the same source, or be in their nature identical. "Likeness" in general may be associated with remarkable difference in particulars, and bodies not to be distinguished from one another in general appearance, may differ widely in power and display marvellous differences as regards the materials formed. In fact many things may be like in a few points which are unimportant, and differ in other points which are of the greatest consequence. Many of the comparisons insisted upon are unsound, and many of the analogies which are stated to exist are analogies which can only be discovered by those who are privileged to spread their own one-sided views, and to ignore the results of observation.

The idea has been for some time generally entertained that lymphatics form an extensive plexus in almost all tissues, and that the connective tissue corpuscles generally, or the tubes extended from them, are in direct continuity with lymphatic vessels. It has been further held by some observers that the so-called wandering cells, and even pus corpuscles in these tissues, really lie within lymphatic tubes and their prolongations. The idea of Virchow that the connective tissue corpuscles were the special seat of development of pus when not formed by epithelium, lent support to the doctrine when the views concerning the origin of lymphatics from connective tissue corpuscles were received. The facts, however, can be explained better without thus piling hypothesis upon hypothesis.

Pus may originate from the bioplasm of tissues, or from white blood corpuscles, &c. The "fibres" of connective tissues are so arranged as to permit the passage of small particles of moving bioplasm amongst them. There is much probably almost diffluent interfibrous fluid which

is continually undergoing change in amount, and which flows in slowly moving streams from and towards the nearest capillary vessels. In this fluid minute particles of bioplasm are always to be found, and if the fluid is rich in nutrient matter, these little bioplasts increase in size and number, and from them pus corpuscles may be formed. In health they are no doubt concerned in the removal of superfluous nutrient matter, and some of them from time to time may find their way into lymphathic spaces, or even through the thin walls of lymphatic or capillary vessels. These particles are so minute that perhaps a hundred would be required to fill the space occupied by a white blood corpuscle.

Of late years the strange doctrine that every pus corpuscle was once a colourless blood corpuscle has found many admirers, notwithstanding the many fatal objections that present themselves to its acceptance. The emigration of fully formed colourless corpuscles from the blood vessels, which is probably only an exceptional phenomenon, and of very occasional occurrence, has been regarded as a constant and essential accompaniment of inflammation, and considered to be absolutely necessary to the process of suppuration, as if the fact in question had been proved beyond doubt in so many instances as to justify us in regarding it as a necessary and constant phenomenon. So many cases of inflammation can be pointed out in which not a white blood corpuscle leaves the vessel, that this latter process so far from being "a necessary part of suppuration" is, perhaps, only an accidental accompaniment of that process. In the inflamed cornea the enlargement of the masses of bioplasm, and the formation of pus from them can be proved; and the same fact can be demonstrated in the epithelium of an inflamed mucous membrane as the bladder or vagina, or in cuticle.

289. Bacteria do not cause Pus.—Then we have the hypothesis which attributes the development of pus to the presence and to the growth and multiplication of bacteria. It has been assumed that the irritation caused by the presence of bacteria or their germs, formed the starting point of the suppurative process. But myriads of bacteria may be present without indicating more than that organic fluids and solids are undergoing decomposition. Instead of the presence of these bodies being a necessary part of a disease, it will probably turn out that they have little or nothing to do with the irritation and morbid process.

290. Properties and Powers of normal Bioplasm, and of the Pus descended from it.—In conclusion, I will venture to offer some remarks upon those properties or powers of bioplasm, by virtue of which the production of pus is rendered possible. Although much of what I shall say will necessarily be rather speculative, the interest of the matter seems to me so great that I cannot pass it entirely over. The bioplasm of tissue, or some other form of bioplasm, being supplied with an increased quantity of pabulum, may give rise to pus. In the process

new powers are acquired. Pus differs in *power* from the bioplasm from which it was derived. It cannot, as far as we know, re-acquire the properties which the original bioplasm possessed. My meaning will be rendered clearer if I adduce an example. The bioplasm of cuticular epithelium may give rise to the peculiar hard material of which the so-called "walls" of the epithelial cell consist, and this same bioplasm of epithelium, if freely supplied with pabulum, may give rise to pus. Bioplasm may therefore *lose* formative power, and become degraded, but it cannot again acquire or regain the power that it has lost. There is, as it were, no return to a high position for living matter which has once suffered degradation, nor can degraded bioplasm, under any circumstances, produce descendants with exalted power.

It must not, however, be inferred that degradation in formative power implies *diminished vitality*. It is a mistake to conclude, as some have done, that *disease* is necessarily associated with diminished vitality. If, indeed, in speaking of the various degrees of vital activity, we refer to the rate of increase of bioplasm, we must admit that in most diseases we have incontrovertible evidence of greatly *increased vital activity*. In inflammation, there is greatly increased vital activity as compared with the healthy state; that is, much more lifeless matter becomes *living* during a corresponding period of time in inflammation.

The formative or developmental endowments of bioplasm are diminished or completely destroyed by its rapid multiplication, so that a mass of bioplasm, which in the course of a considerable time undergoes comparatively slow change, would give rise to descendants which might be concerned in the development of the highest and most complex tissues, if placed under conditions favourable to its too rapid growth would absorb much more nutrient matter in a given time, and produce descendants much more quickly. But, however long this rapid multiplication might continue, not one of the resulting masses could form the characteristic normal tissue. Too rapid increase is associated with degradation in power. And in man and the higher animals, and plants, if textures grow too quickly, the perfection of the tissues formed is marred, and the period of their duration is of necessity reduced.

Ordinary pus, then, may readily be produced if the nutrition of the bioplasm of a normal elementary part of any tissue be increased. Under certain specific conditions which we are not yet acquainted with, pus with peculiar and specific properties or powers, is formed, and this exhibits a far greater vital activity and is less easily destroyed than the first. In this way, indeed, particles of bioplasm come into existence which exert a peculiar influence, giving rise to a definite series of changes quite peculiar and characteristic of that particular form of bioplasm only. Such particles are, indeed, "disease germs," which I shall now proceed to consider.

On the Poison, Contagium, or Virus of Contagious Fevers.

291. General Nature of the Poison.—The general views which, after numerous investigations upon the virus of different contagious fevers, I have been led to adopt concerning the particles of matter which constitute the active agents in transmitting the disease—the *actual poison*, *contagium* or *contagion*—have been incidentally alluded to in preceding sections. It was shown in the last page, that a change in the conditions under which the normal bioplasm of the body lived, might *determine an alteration in the rate of its multiplication, as well as an alteration in the properties and powers of the living particles produced.* From bioplasm thus placed, it has been shown, pus may result. This pus bioplasm not only grows and multiplies with far greater rapidity than the living matter from which it came, but the particles, unlike those from which they came, may be removed long distances from the seat of their production, without their vitality being destroyed or their vital powers impaired. Arguing from the data afforded by investigations upon certain forms of pus production, I believe I am fully justified in inferring that the very rapid increase of certain forms of bioplasm, perhaps of those constituting the living particles of the blood and of the lymph under conditions which have not yet been investigated, is followed after a time by the origin of living particles, having a power of resisting the influence of external circumstances far stronger than is possessed by pus, and a power of increase and multiplication under special circumstances, compared with which the growth and multiplication of pus are but slow changes. In my report on the Cattle Plague, some observations bearing upon this matter are recorded, and these, as it seems to me, point to the general conclusion that the virus or contagium of contagious fevers, like pus, has originally been produced by direct descent from some form of normal bioplasm of the body. Nay, so very like are certain contagious particles to the bioplasm which results in the course of inflammation that they are generally considered to be pus-corpuscles. Unquestionably, they grow and multiply, like pus. In my work on "Disease Germs," I have directed attention to the case of the development of a very virulent living poison in peritonitis. Here it may be truly said that we almost see the birth of a contagious virus, capable of infecting another organism, and of being carried from place to place, the particles of which may retain their vitality for a considerable period of time, in air, in water, and in other fluids, upon the clothes, and about the person.

The infinitesimal quantity of such contagious virus that may adhere

to the cuticle of the fingers, after careful washing, is sufficient to spread disease. Of this fact we have evidence only too distinct, in some terrible examples of the class of cases included under the head of puerperal fevers. The indistinguishable and not to be detected trace of the poison of erysipelas removed by a touch, or of peritonitis adhering to the cuticle, in spite of efforts to remove it, has been conveyed to the organism of a puerperal woman. The living particles have grown and multiplied therein, and have caused the death of the patient.

So virulent are some of these poisons, and so strongly do they resist the destructive influence of external conditions, that they not only retain their vitality, but may actually grow and multiply in situations and under circumstances having little in common with the seat of their production. Such poisons, it need scarcely be said, are not easily extirpated by us. But if every particle of a given contagium were destroyed, there is no reason for concluding that a new contagium, nearly allied to it, would never again be generated. On the contrary, it may be regarded as almost certain that, if fevers of certain familiar types ceased to prevail, fevers of allied characters or of markedly different types would be developed. Nay, I cannot think that it is in any degree more probable that specific contagious fevers will cease to afflict man and the higher animals than that the feverish state will be abolished or prevented. Fevers will probably last as long as the human race shall exist, and from time to time new fevers will be generated and old ones will disappear.

The reader will have already discovered that the views to which I have been led upon the nature of contagion are different from those generally entertained at this time. It is desirable that, before I refer more particularly to the character of diseased germs, the doctrines so widely different from my own should be briefly recounted, and the points which, as it seems to me, render them defective or untenable, alluded to.

292. Fungus Germ Theory of Disease.—The notion that disease germs consist of very low forms of microscopic fungi has been more than once advanced and abandoned, but during the last few years this theory has been revived, and has received the sanction and warm support of authority. Like some more fanciful hypotheses, it has gained energetic advocates who happen to have facilities for making this or any other doctrine, for a time, very popular. Views which do not rest upon such ascertained facts not unfrequently become widely known, irrespective of their merits, and before the points for and against them have even been considered. A short-lived notoriety seems only to be desired, and this in our day is easily obtained.

293. Zymotic Diseases.—For fifty years or more we have been taught that fevers exhibit an analogy to fermentations, and it has been

assumed by many that a sort of fermentation is excited in the blood of the victims by the poison of the contagious disease, in somewhat the same way as the wort is caused to ferment by the presence of the yeast fungus.

Diseases of the kind referred to, and known to be dependent upon a poison which increased and multiplied in the affected organism, have been termed "zymotic," and have long been included under this head in the reports of the Registrar-General. But the meaning of this word "zymotic" has, like many other scientific terms, undergone change. The word used to be applied only to processes dependent upon the presence of a tangible substance afterwards discovered to be a vegetable organism, and termed a ferment. Then its application was extended so as to embrace the phenomena which occur in man and the higher animals, in fevers which are certainly contagious. But recently, even tubercular disease has been regarded by Mr. Simon as a "chronic zymotic process," the contagium of which, he remarks, appears to be "the common septic ferment." Now, by like reasoning, we might include every pathological change characterised by the growth and multiplication of minute living particles in the class of "zymotic diseases." The formation of pus and cancer would be a zymotic change. Nay, the phenomena occurring during the development of every part of the body, would have to be included among zymotic processes. But neither tubercle nor cancer nor pus, nor, I venture to think, the contagious matter of contagious diseases belong to the vegetable world, nor are they allied to such bodies as fungi, save that, like the latter, they are alive and, according to the fancies of our day, have enjoyed the privilege of springing from a common ancestor. All consist of bioplasm or living matter, derived, not from fungi or from any vegetable organism whatever, but from man himself or from some of the higher animals. All living matter is self-multiplying, and every form of development and growth might in this sense be spoken of as "zymotic," but, in that case, the word would come to be but another name denoting vital phenomena or actions. Is it not, therefore, desirable that the word "zymotic" should be restricted to actual fermentations, depending upon the ascertained presence of vegetable organism? Fevers and other diseases should not be included in this category, at least until the "ferment" shall have been demonstrated to be a vegetable organism, and its characters and properties defined with as much precision and certainty as the sugar fungus, penicillium glaucum, and other fungi.

294. Schizomycetes.—The particles supposed to possess disease-causing properties have been termed *Schizomycetes*, a class distinct from fungi, but including the lowest forms of life known, such as the organisms which have been called zooglæa, bacteria, microzymes, vibrios, and some others, also known by several names. In my report on the cattle plague, and for some years before this was published, I described

the characters of the minute solid particles which I believed to be the active agents in the transmission of disease; and in 1863 I gave figures of the appearances of vaccine particles under very high magnifying powers. To me it seemed impossible to regard these as bacteria, or to mistake them for those particles. Ferdinand Cohn, however, in 1872 (Virchow's "Archiv.," band lv, p. 229, 1872), stated that the solid particles of vaccine lymph were bacteria. I can only say that I have many times examined fresh vaccine lymph, and have never seen any particles which I should consider to be bacteria. The particles I have seen are not of a regular shape—neither round nor oval, nor dumb-bell shaped, nor is there any appearance of an envelope or of transparent matter outside the envelope, as one finds in the case of all bacteria. The figures representing the particles of vaccine as mere circular outlines, are altogether incorrect, and convey a most erroneous idea of their appearance. I have never been able to discover a bacterium in fresh vaccine lymph. It is unfortunate that Sanderson and other authorities in this country receive the views of Cohn and other German observers, as if they must be devoid of error, and as if these authors were infallible.

Although very early in this field of enquiry, my observations have been scarcely noticed by some who have written on the subject; and in some memoirs my work has evidently been intentionally ignored. An English observer must expect this at the hands of some of his countrymen, especially those belonging to certain schools of thought, but at the same time it is right that attention should be called to the circumstance. Some writers displaying more ingenuity than candour, seem preparing to extend such terms as microzymes, micrococci, to the minute particles of living matter which I have figured, and have shown to exist in the blood and in other nutrient juices of man and the higher animals, as well as in the moving matter of certain vegetable cells,—particles which are composed of bioplasm, and endowed with properties very different from those possessed by the bodies in question. I have shown that minute particles of pus bioplasm can be derived by direct descent from the normal bioplasm of the body, and not from microzymes or micrococci, or any vegetable or other living bodies which come from definite organisms of the lowest kind, and produce the like. These last have no relation, as regards origin or descent, with the bioplasm of the higher organisms in the tissues and fluids of which in a healthy and morbid state they may live and multiply, and upon the constituents of which they feed.

295. The Bacterium Hypothesis of Contagium.—The idea that the different forms of contagium consisted of living particles of some of the microscopic fungi, has of late been given up for the nearly allied doctrine that the contagium particles are bacteria, different species of

bacteria representing the several contagia. Instead of being convinced by the fact and arguments advanced that such a conclusion is justified, I cannot but think that the doctrine that contagious diseases are caused by bacteria would have been pronounced untenable by any unprejudiced observer at any time during the last twenty years. Some of the leading scientific minds of our day seem to act as if their main object was to make a good scientific case. No wonder we find ingeniously-elaborated arguments which have been highly popular fall to the ground as soon as the artificial support they have received is withdrawn. The public is at once convinced by the declaration of an authority. Contagion accordingly takes the form of a microscopic fungus, or a chemical compound, or it is due to a condition of the air, to electricity, or to a want of electricity, as authority may dictate. Any one suggesting defects in the facts or arguments may be accused of attempting to arrest the elucidation of truth, and of interfering with the progress of knowledge.

Of all the hypotheses yet advanced, the recently revived bacterium hypothesis has unquestionably gained the support of the greatest number. It may boast of being accepted by many scientific men—physiologists, physicists, sanitary authorities of great eminence, physicians and surgeons. Such energy has been displayed by its advocates, that few readers can be unacquainted with its terms. And yet I doubt whether an hypothesis could be pointed out which is less justified by the facts of observation upon which it is professedly founded. Because micrococci are found in the tissues in erysipelatous inflammation, we are expected to accept the conclusion at once that the disease is caused by the growth and multiplication of these particles. Because bacteria have been found in vaccine lymph and small-pox lymph, we are to accept the inference that the wonderful properties of these fluids depend upon the organisms above-mentioned.*

Not a few authorities, more distinguished by ingenuity than candour, leave the reader in doubt concerning their real opinion. With an affectation of cautious reserve, they so frame their statements that at a future time they may either claim to have advocated or to have dissented from the bacterium hypothesis. It may be found convenient by and bye to suggest that by the term "microzyme" it was only intended to convey the idea of a very minute particle of living matter—not necessarily a particle in any way related to a bacterium, or a fungus of any

* In his last report, "new series," No. III, page 30, "Reports of the Medical Officer of the Privy Council, &c., 1874," Dr. Sanderson again refers to the discovery by him in vaccine lymph of minute bodies, which he calls micrococci, and which he says are usually present (12th Report, 1870). He says Professor Klebs described them about the same time, and that two years afterwards the subject was treated with more completeness by Ferdinand Cohn, whose observations "related both to vaccine lymph and variolous lymph, and led to the interesting discovery that the organisms which I (Dr. Sanderson) had described in the former are also found in the latter." I have had

kind. Affectation of excessive caution in giving an opinion upon a scientific question may shield a philosopher suffering from confusion of thought, but all the interest connected with the discussion of this subject disappears if an authority, after a full study of the matter, tells people that the poison of a contagious fever may be a bacterium, or something allied to a bacterium, or something not allied to a bacterium. Those who can get no farther than this, might at least condescend to acknowledge the observations of other English observers.

With regard to this bacterium hypothesis, I will now call the reader's attention to the following facts and considerations :—The presence of these living organisms in the tissues and fluids of the body is not pecu-

Dr. Sanderson's wood-cut copied below. Such bodies as those delineated are not to be found in clean fresh vaccine lymph, or in small-pox lymph. Such figures are calculated to mislead, and it is unfortunate that they should find a place in an authoritative Government report. The advance of science may be much retarded if Government officials are permitted to publish in departmental blue-books scientific doctrines in which they are interested, and to ignore facts and observations which happen to be opposed to their pet theory. I shall further express the opinion that if the researches and observations of German authors are to be published at the public expense, it is not right that the results of the labours of men in this country should be omitted. The compiler should be instructed to draw up a careful report of the observations of all those who are known to have published memoirs upon the subject, and he should be informed that he is not to select only those which he approved and the special results which favoured his own views. The public could draw their own conclusions if they were permitted to study the different statements. They want a fair and impartial statement of what has been done by different persons, not a report of researches in favour of one particular doctrine. It may fairly be doubted whether by the publication of lengthy and often confused accounts of foreign observations, much that is of real advantage in advancing our knowledge is gained. Such reports are not calculated to encourage original thought or original enquiry. They are likely to be regarded with most respect by persons who do not read them. Moreover it is impossible for any one to form a correct notion of the thing seen by a man from description alone. The drawings of different observers ought to be placed side by side, though this would be objected to on the score of expense. After reading through many pages of matter selected as important, we sometimes find the consciententious commentator himself obliged to remark that the question still remains open, or that the experiments are insufficient.

"Groups of particles in fresh vaccine, drawn with the camera lucida, and magnified 500 diam." Copied from Dr. Sanderson's figure.—Twelfth Report 1870, page 232.

If the commentator particularly desires to make a good case, he picks out certain observations and reports these with a prolixity that is tedious. Many pages are filled at great cost, and an impartial reader discovers that nothing definite has been gained. A writer may desire to appear very learned to the unlearned, to publish the views of friends, or to confuse people for the purpose of deterring them from troubling themselves with the matter at all.

liar to contagious diseases. Bacteria and their germs have been found in numbers in cases characterised neither by fever not by specific symptoms, nor by blood-poisoning, nor by being propagated by contagion. Nay, bacteria grow and multiply, and in all organisms and in all parts of organisms, so very soon after death either of a part or of the whole body has occurred as to render it almost certain that their germs must have existed in the organism before death. These minute organisms, considered to be the causative agents of specific fevers, exhibit similar characters in widely dissimilar diseases. They live and multiply under many different conditions, and consume healthy as well as morbid matters in a state of decay. Such are some of the arguments against this now favourite hypothesis upon the nature of contagium. But on the other hand, it must be admitted that the discovery of a nearly allied organism in the blood in one particular fever, and in that only, is a very remarkable circumstance. Dr. Obermeier, in 1867, found numerous specimens of *spirilla* in the blood of persons suffering from relapsing fever. The organisms disappeared when the fever passed off, and were found again when the relapse occurred. These observations were confirmed by Engel, Weigert, Litten, and others. "Reports of the Medical Officer of the Privy Council," New Series. No. III., page 42.

The organisms are found during the paroxysm, but usually disappear after the temperature has risen. They are never found when the feverishness ceases. It has, of course, been concluded that the spirilla organisms constitute the real contagium of relapsing fever, but such an inference cannot be justified by the facts so far ascertained.

Lister's well-known researches in 1867 led him to conclude that the process of suppuration was due to the direct action upon the wound of living organisms derived from without. But there is little doubt that the idea that suppuration is caused by bacteria getting into a wound from the air is erroneous. It seems indeed to be sufficiently disproved by the fact that suppuration often occurs although no bacteria can be detected. His deduction that in the treatment of wounds bacteria ought to be excluded by carbolic acid vapour through which they cannot pass alive cannot be sustained. That the carbolic acid is of great use I am convinced, but the explanation given by Lister is probably not the true one. His practice is doubtless excellent, but the data upon which it is based may nevertheless be faulty. It seems very probable that the favourable results of this method of treatment are due not to the destruction of the bacteria, but to the direct influence of the carbolic acid upon the wound. Carbolic acid interferes with the rapid growth and multiplication of bioplasm, which results after a time in the formation of pus corpuscles.—(See page 248).

296. The Bacterium and the Bacterium Germ.—There is a great

difference between the *bacterium* and the minute *germ* particle from which it springs. This little body is often less than the $\frac{1}{100000}$ of an inch in diameter, and there are probably few living things in existence which will retain their vitality for so long a period under such very different adverse circumstances. It resists extreme cold and great heat, nor is it destroyed by dryness. The observer must therefore draw a distinction between the quickly growing, multiplying change effecting the bacterium which flourishes under certain circumstances, and grows and multiplies in certain fluids, and the passive and more minute germ-particle from which bacteria originate under particular conditions, These germ-particles, each one probably protected by an envelope, may remain dormant, passive, and with little change for a great length of time. Such particles exist everywhere, and retain their life under conditions which would not only infallibly destroy the growing, multiplying bacteria, but every other living organism known to us.

In considering the merits of the hypothesis that bacteria are the agents concerned in propagating contagious fevers, we must bear in mind their universal distribution. Whether disease exists or not, bacteria are present, and there are few things that we eat and drink that do not contain multitudes of them.

In internal parts of the bodies of man and animals, in their tissues, and in their blood, these little germs exist and probably multiply, but very slowly, their life-changes being in abeyance as long as the bioplasm of the body with its higher life prevails; but when this succumbs, the bacterium-germ bursts its bonds, and its descendants, soon numbering millions of bacteria, assert their sway to the detriment of the part. There is no place in the world from which bacteria-germs are known to be absent, nor is there an organism, high or low, great or small, vegetable or animal, healthy or diseased, in which the bacterium-germ does not exist. Undoubtedly it may remain passive for years, perhaps for centuries, and in blazing light and impenetrable darkness, in tropical heat and in polar cold; but, nevertheless, the germs live, and the instant the circumstances become favourable to their development they germinate. Nay, it is possible that even in its passive state the bacterium-germ may slowly multiply in media not favouring its rapid increase, but nevertheless not incompatible with its slow growth.

It appears that a temperature high enough to destroy every other known form of life will not kill the bacterium-germ at least at a certain stage of its existence. The germ manifests resisting powers which the fully-developed bacterium does not possess. This, however, is not peculiar to the bacterium—a similar fact being noticed with regard to many higher forms of existence. Messrs. Dallinger and Drysdale have conclusively proved that the germs of certain monads will retain their vitality at a temperature of 148° cent., although the monads themselves

were destroyed at 80° cent. The germs in question being so minute as to be invisible under a magnifying power of 2,000 diameters, their powers of resisting the influence of heat can scarcely be attributed to the thin envelope in which the germs may be enclosed, but it must rather be referred to some property inherent in the living particle itself. At the same time the fact must be borne in mind, that these, like all other living forms, are infallibly killed by the *prolonged action* of a temperature many degrees below 148° cent. Whether the disturbance in the arrangement of the component atoms of the living particles is prevented by some influence of the protecting medium in which the bioplasm lies, or whether the forces which determine the positions of the atoms can only be overcome by slow degrees—one atom after another, as it were—cannot be conclusively determined.

297. Further Arguments against the Bacterium Hypothesis.—That bacteria are not in themselves hurtful has been proved most conclusively. Many things we eat contain them in countless multitudes. In the alimentary canal of infants suffering from a little stomach derangement bacteria are often present in vast numbers, and in some of these cases not a particle of the contents could be found at any point between the mouth and the anus that was not teeming with bacteria. Many of the secretions may contain them without any perceptible injury to the health, while hosts of them are invariably present in the fluids, and in and about the superficial cells of the mucous membrane of the mouth of all persons, even in the most vigorous health.

Before we accept the doctrine that contagium consists of bacteria, we must remember that in the most virulent animal poisons bacteria are not to be found until the virulence and specificity of these poisons have been destroyed by commencing decomposition. Then, of course, millions of bacteria exist. In all dead and changing animal and vegetable matter, healthy and morbid, bacteria grow and multiply. There is no morbid or contagious matter, however inveterate, that bacteria will not appropriate, and at the expense of which they will not live and grow; and yet it is maintained that these organisms which can assimilate poisonous contagium are themselves the contagium! Is there a doctrine of modern times that can be fairly said to be less supported by observation and experiment than this favourite bacterium hypothesis of disease?

But already some of the warmest supporters of the hypothesis are beginning to modify their views, and suggest that even if bacteria are not themselves contagia, they may be the carriers of contagion. It might be further suggested that bacteria may be instrumental in preparing the ground for the reception of contagion; but there is this difficulty:—so far from bacteria growing and multiplying *before* the contagium, they succeed it. There is the same objection to the acceptance

of the doctrine that bacteria generate the contagium. Some may maintain that the contagium generates them, others that bacteria lie dormant in contagium, or the latter in them. The *bacterium* may be a *bacterium contagiosum*, diseased in consequence of having fed upon morbid poisons, or the secretion formed by the *bacterium contagiosum* may be the poisonous agent, and not the bacterium itself. Such conjectures might be multiplied *ad infinitum*, with the only result of boring readers, and exciting contempt for the hypothesis-mongers on the part of sensible people.

It is extraordinary that the bacterium-germ hypothesis should have won so many converts. Those who accept it, of course assume different species of bacteria, each one corresponding to a specific disease; but up to this time no contagious disease whatever has been proved to be due to any bacterium, or to a bacterium-germ, while, as has been already pointed out, many potent animal poisons when most active are destitute, or nearly so, of bacteria. Indeed, the virulence of the virus decreases as the bacteria in it increase in number. Again, animal fluids, which contain multitudes of bacteria, do not produce specific contagious diseases. But rather than admit that the bacteria present in specific contagious poisons are concomitants, and have nothing to do with the specific contagious property, many prefer the doctrine that every specific virus consists of bacteria.

298. Another Hypothesis.—But very different hypotheses, though perhaps as unreasonable, have their advocates and supporters. It is maintained by some that contagium is not living at all, but consists of lifeless particles, which cause chemical changes in the fluids and tissues of the body. But this view need not be discussed here.

As regards Dr. Bastian's views upon contagion, I shall content myself by giving, in his own words, a sort of interrogative summing up with which he concludes the last of his voluminous writings on Archebiosis:—"May we not say that chemical changes established in some one tissue, or in many, by dint of altered blood and other secondary processes, spread so as to be initiated also in previously sound parts; and that thus throughout the body, or in some special regions of it, living tissue endowed with peculiar and poisonous properties, or complex alkaloidal compounds, may be engendered in enormous quantities, some of which may be thrown off from this or that surface, and act after the fashion of 'contagia' generally?"* The writer considers that his conclusions, as expressed in the above sentence, belong "to the region of fact and warranted inference"!

299. The Contagious Bioplast.—The contagious bioplast is a living particle of extreme minuteness, in most cases less than the $\frac{1}{100000}$th of an inch in diameter. It is invariably colourless, and often so clear and structureless that it is only with difficulty discovered in the fluid medium

* "The Monthly Microscopical Journal," September 1st, 1875, page 140.

in which it is suspended. Such a particle may be easily transferred from an infected to an uninfected organism. It may be carried a long distance from its source of origin without losing its vitality, and even after having passed through different media, and after having been exposed to considerable changes of temperature, it may excite in the invaded organism a series of changes resembling, even in very minute particulars, the phenomena which characterised its presence in the organism from which it was immediately derived. Nay, some contagious bioplasts will grow and multiply in certain animal fluids—milk, for example,—and thus the particles may increase in number out of the body.

Each kind of contagious bioplasm manifests its own specific actions, and only these. The bioplasm of small-pox will produce small-pox, not typhus, nor measles, nor pyæmia, &c., nor will any one of the latter forms of bioplasm give rise to small-pox.

The facts and arguments I have advanced prove, I think, as regards the virus of contagious fevers :—first, that it is living, growing bioplasm, and derived from bioplasm ; secondly, that the particles have not been generated in the organism of the infected animal, but have resulted from the multiplication of one or more particles like themselves, somehow introduced into the body ; thirdly, that the contagious bioplasm particles grow and multiply in the blood and some other fluids of the animal ; fourthly, that the bioplasts are sufficiently minute to readily pass through the walls of the capillaries, and may multiply even in the interstices of the tissues, or between the tissue-elements and epithelial cells ; fifthly, that they will live under many different conditions—growing at the expense of various tissue-elements, and retaining their vitality though the bioplasm of the normal textures, after growing and multiplying to a great extent, shall have ceased to exist ; and, lastly, like the pus bioplast, all forms of contagious bioplasm that invade man and the higher animals were originally derived by direct descent from the bioplasm of the body of man or animal, and are not of the nature of bacteria or akin to any low form of vegetable organism.

The contagious bioplasm I have described is not soluble in water, as many believe the virus of contagious diseases to be. That Klebs should state, even with respect to the tubercular virus, that "it is soluble in water," seems to show how very vague are the general views still entertained upon the nature, not only of contagious virus, but concerning what goes on in some broad pathological changes. No form of living matter is soluble. Solubility implies that matter is not in a living state. Death must precede solution ; for, indeed, that which is dissolved cannot be living matter, but may have resulted from the death of living matter.

I have given some figures of the minute particles of contagious bioplasm in pl. XLV, figs. 3, 7, 8. In fig. 8 some bioplasts of vaccine are represented as they appeared when magnified five thousand times.

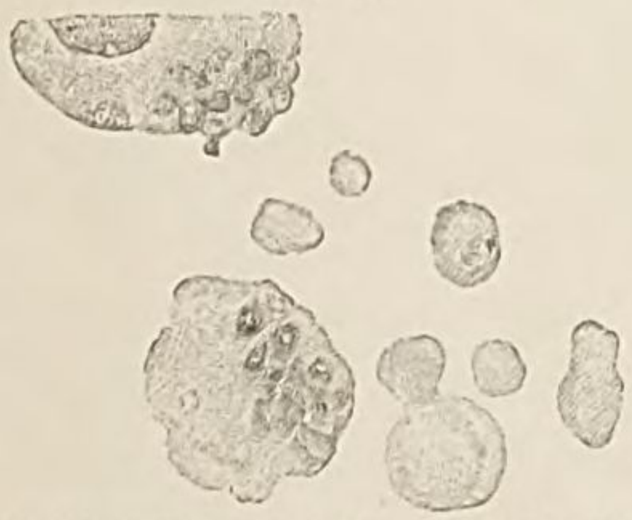
Fig. 1.

Fibrous tissue of corium or true skin from the softened part of a papule from the udder of a cow suffering from cattle plague. The spaces between the fibres are completely occupied with contagious bioplasm (disease germs) growing and multiplying rapidly. × 215. p. 320.

Fig. 2.

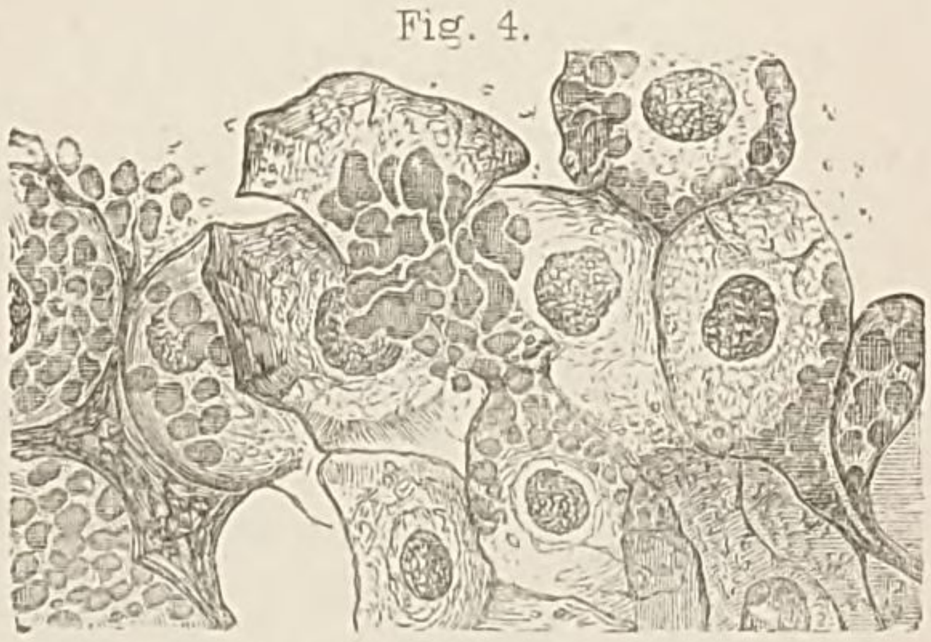
A part of Fig. 1, magnified 700. The particles of contagious bioplasm are dividing and subdividing rapidly and invading the bundles of fibrous tissue.

Fig. 3.



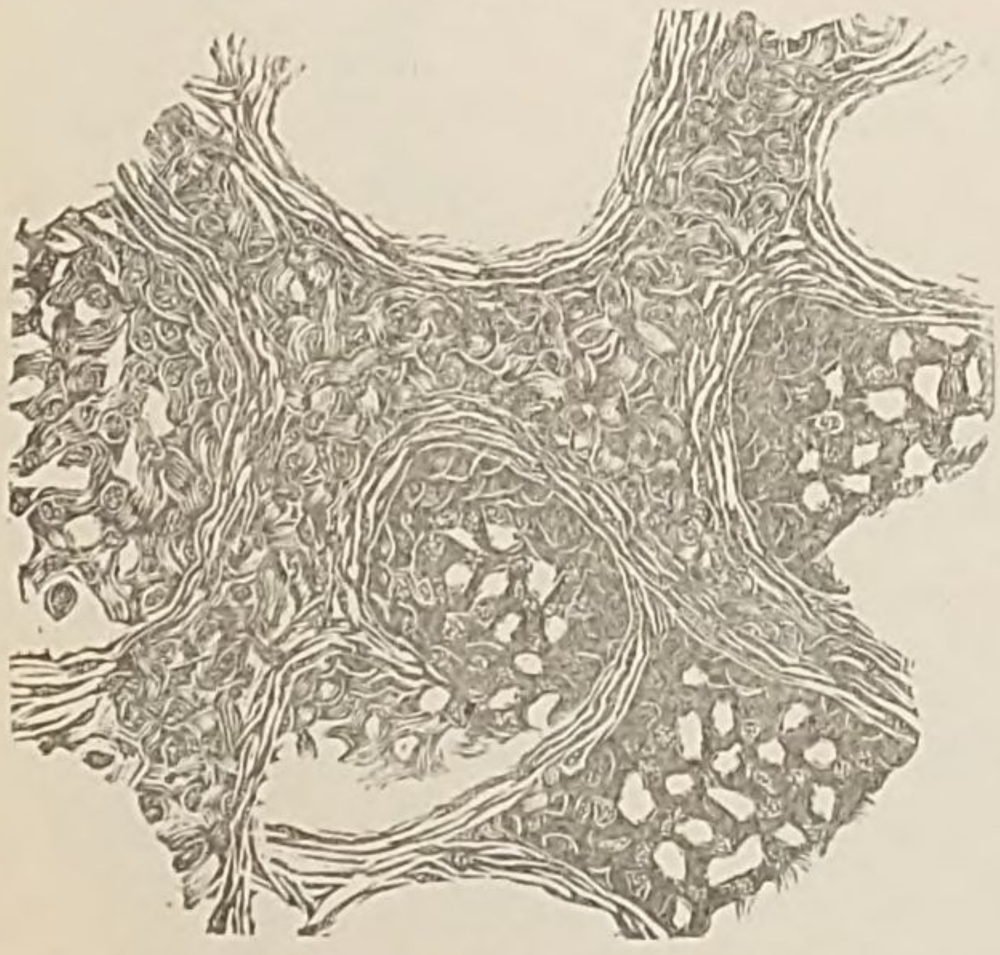
Most minute particles visible of living bioplasm in the milk cattle plague. These are probably the actual contagious bioplasts of that disease. × 2800. p. 320.

Fig. 4.



Cuticular cells under the scab. Eruption on mamma, cattle plague, showing the cells invaded by the growth and rapid multiplication of the contagious bioplasts, × 700. p. 320.

Fig. 5.



Air cells from a healthy human being, in which the capillaries had been injected It is in the bioplasts seen in all parts of the specimen that the earliest changes in the development of tubercle occur. ×130.

Fig. 6.

Bacteria and vibriones from ovine smallpox vesicle. × 1300. From PROF. BROWN, "Veterinarian," 1868. p. 667.

Fig. 7.

Minute particles of contagious bioplasm from the fibrous tissue of the skin. From the specimen represented in Figs 1 and 2. × 1700. p. 320.

Fig. 8.

Living particles from vaccine lymph. × 5000. p. 320.

$\frac{1}{1000}$th of an Inch ⌊___⌋ × 215.

" " ⌊_____⌋ × 1700.

[To face page 320

If but one of these gained entrance into an organism prepared or adapted for its reception, it would soon absorb nutrient fluid, grow and multiply, and form a small collection,—some particles of which, gaining access to the lymph, or blood, or both fluids, would be carried to more distant parts, and become impacted in some minute vessel. The process would be repeated, and in this way every part of the organism would soon become infected. From collections of poison bioplasts formed in a capillary or lymphatic, some minute particles would easily pass through minute interspaces, and gradually make their way, moving as a mucus-corpuscle, or a white blood or pus corpuscle moves, and at length perhaps appear upon the free surface of the body. Some might be carried away in currents of air, others, clinging to external objects, might remain some time, and be brought into contact with and infect an organism. In many ways that need not be recounted the bioplasm particles may be carried from the infected organism, and the disease spread far and wide.

The views above given concerning the nature and origin of contagious bioplasm, and the facts upon which they are based, are recorded in my report to the Cattle Plague Commissioners, who, in their summing up, informed the public that I had discovered "granular matter." By this remark they publicly, though I believe unintentionally, condemned my work, for who would care to study the memoir of an author who had made such a remarkable contribution to our knowledge as the discovery of "granular matter?" It is by ignoring that most important matter of the body, *bioplasm*, or living matter, or by calling it granular matter, that authorities succeed in diverting the public mind from the study of its various forms, and from the contemplation of its wonderful powers in simple and complex organisms, and in the healthy and morbid state. But man himself at an early period of his life is but "granular matter" if structureless living matter is to be so styled.

TUBERCLE.

In a work like the present, a very full discussion concerning the nature of tubercle would be out of place, but it is not possible to treat of the subject in any way useful to the student without referring to some of the theories now generally entertained. Unfortunately many of these are very complicated, and the ideas adopted often not clear. New views have too often been grafted, as it were, upon old doctrines, and sad confusion is the result; besides, so-called "observations," but which are in truth mere speculations, have been appealed to, and however worthless the conclusions may have been, hostile criticism has not only excited anger, but has led to more observations of the same sort being published, to confirm the first. In this, as in many

other departments of scientific investigation, we find multitudes of observations associated with but little real progress.

Among the most interesting of the modern views concerning the nature of tubercle, attention should be directed to the following :—

Schrœder van der Kolk held that tubercle resulted from a change taking place in the ordinary epithelium of the pulmonary air cells, and this, or a view closely allied to it, has been entertained by a great number of authorities. Of the existence of the epithelium in question, however, at least in the healthy state, there is some doubt, and, supposing its existence be admitted, there will be found great difference of opinion concerning its exact nature.

Some observers have convinced themselves that all true tubercular matter may be known by its *caseous* or *cheesy character*. Others affirm tubercle to be dead matter, the mere products of decay, but perhaps due to a special disintegrative process. One authority asserts one "cell" to be distinctive of tubercle, another a different sort of cell. Huge cells, it has been recently said, are characteristic of tubercle, although in many well-developed examples not a large cell can be seen. Then these cells are supposed to be formed by coalescence—a doctrine which indicates some strange disturbance of the imaginative faculty, for the idea of epithelial cells enlarging and then becoming "fused together to one large mass of granular protoplasm" is nebulously conjectural. There is no evidence that epithelial cells ever become fused together, and the idea that any form of protoplasm could be produced by such a process is simply untenable.

It has been suggested, again, that tubercle, like many another obscure disease-inducing particle, consists of a very low vegetable fungus, or some other special organism perhaps allied to bacteria. "Observations" are not wanting to bolster up this idea, which just now of course finds many advocates. Any one, however, who has examined tubercle at different periods of growth and development will be satisfied that it is not in any way allied to any bacteroid body, or to any fungus. Some, dissatisfied with every definition, explain tubercle away, and affirm that "tubercle" only exists in the imagination.

Several writers in England have recently accepted conclusions arrived at by German observers tending to prove a far more intimate relationship between lymph corpuscles and tubercle than had before been suspected to exist. But, as in many other cases, we find an idea more definitely stated and more fully elaborated than the facts upon which it is based warrant. Believing firmly in the *tendency* of thought, new observers repeat observations, and enthusiastically confirm the discoveries originally advanced, and extend them farther than their originator. Difficulties are passed over, objections overruled, and very soon the views gather such force that objections cease to be listened to, and the doctrine

is expounded more in the style of dogma, and promulgated as if it rested upon infallible authority, instead of being submitted as an inference based upon the results of observation and experiment, and liable to be modified or upset by new observations.

For years it has been known that lymphatics were freely distributed beneath the pleura, and that many existed in the sub-mucous tissue of the respiratory passages, but now we are asked to assume, what has not been demonstrated, a plexus of lymphatics around every pulmonary air-cell; and we are to believe further, that in the process of tuberculization lymphatics and lymph corpuscles perform an essential part. Nay, we are asked to regard the air-cells of the lungs as lymph-spaces filled with air instead of with lymph (Buhl). May not the wind-pipe after all be but a modified vessel?

More recently "giant cells" have been announced as peculiar structural elements of tubercle, and these giant cells have been formed, it is said, by the fusing together of the cells of the air-cells of the lung. Others, as already remarked, describe a mass of protoplasm resulting from the *coalescence of cells*, but this hypothesis is opposed to many facts that are well known. Many of the most recent "explanations" of the formation of tubercle could only have been advanced by observers who had had very little practical knowledge of minute pathological changes, and were but imperfectly acquainted with the practical details of minute investigation. Networks of nucleated cells are called into existence, when required, and lymphoid corpuscles discovered in their meshes. Cheesy metamorphosis and fibroid and other metamorphoses with various degenerations, assist in the construction of a confused and artificial system, bolstered up by contradictory statements and facts of the imagination. But what is still more remarkable, is that we are asked by one authority, who also appeals to "facts," to believe that this substance tubercle, which grows and multiplies, *is capable of being dissolved in water!* It seems to me extraordinary that so recently as 1870 Klebs should have affirmed that "the tubercular virus was soluble in water."—(Virchow's Archiv. f. Path. Anatomie, 1870, Bd. 49, p. 291.) He might as well affirm that a living white blood corpuscle, an amœba, a bacterium, nay, an embryo was soluble in water. It is certain that matter that lives cannot be soluble—solubility belongs to that which is non-living only.

I showed many years ago that tubercle, at least during the early and most important period of its existence, when all those serious changes resulting in hopeless damage to the lung tissue are initiated, consists of small particles of bioplasm about the size of a red blood corpuscle, capable, like other bioplasts, of growth and multiplication. Tubercle bioplasm is as distinctly proved as is that of pus. It has special characteristics. It passes through certain phases of growth. Like every

other form of living matter, it dies, and the products resulting from its death are peculiar, varying, however, within certain limits, according to the circumstances under which death takes place and the period of its development at which death may be occasioned.*

300. Examination of Tubercle.—Specimens of tubercle may be taken from the lung itself, or from other organs in certain cases of tubercular disease. The most typical examples of tubercle in an early stage of development are to be obtained from the lungs of growing children, who have been victims of acute pulmonary tuberculosis. One of the minute granulations may be snipped off and moistened with serum, or a solution containing 1 per cent. of chloride of sodium, or placed in a little vitreous humour. If, after gently tearing with needles, the thin glass be applied, and the specimen slightly pressed, many separate particles, "tubercle corpuscles," epithelial cells, blood cor-

* I shall venture to remark that, in my judgment, the pathological views in connection with this subject lately introduced are not justified. The "facts" upon which some conclusions have been based have been wrongly interpreted, or are unreliable. Far too much has been assumed, and instead of the observer's imagination being controlled by his observations, we have much reason to fear that the facts adduced in some memoirs as the results of observation, have been established for the purpose of supporting the author's preconceived and it may be purely theoretical assumptions.

The following extract is from a review in the "Lancet," of November 13, 1875, and gives a summary of conclusions which have been favourably received, and probably accepted, by many who have not the time or inclination to consider carefully the character of the phenomena described. There are many statements to which some exception may be taken, while the general account given in the extract seems to have been founded upon erroneous interpretation, if not upon defective investigation.

"The author" (Dr. Klein) "accepts Buhl's description of a miliary tubercle in man. Young tubercles possess a matrix of peripheral connective tissue arranged circularly, and a homogeneous reticulum without nuclei, enclosing cells smaller than lymph-corpuscles, multinucleated giant cells, and epithelial cells. Buhl considers that all cases of acute miliary tuberculosis are caused by a disseminated catarrhal pneumonia, and that this process differs from genuine desquamative pneumonia only in the presence of giant cells among the germinating epithelium of the alveoli. From an examination of the lungs of seven children who died from undoubted acute miliary tuberculosis, Klein has come to the conclusion that these cases have *at the beginning only* the character of desquamative pneumonia. In the earliest stages the nodules correspond to groups of alveoli and infundibula, distended by a fibrinous material containing granules and small cells. This exudation disappears, and is succeeded by groups of cells, or by one multinuclear giant cell, both derived from the alveolar epithelium. The interalveolar tissue is thickened and transformed into a retiform tissue, which contains lymphoid corpuscles. The giant cells give rise to nucleated cells, which increase the retiform tissue, and also to cells resembling lymphoid corpuscles lying in the meshes of that tissue. Commencing from the centre of the tubercle, the giant cell, as well as the surrounding tissue (after conversion into a dense fibrillar substance) degenerates, and becomes a hard, firm, and débris-like mass. After these necrotic changes have begun, the blood-vessels are surrounded with a true adenoid tissue analogous to the perivascular cords above described, and spherical collections of lymphatic tissue appear in the adventitia of the bronchi."—"Lancet," November 13, 1875.

puscles, and other anatomical elements will be detected. The so-called "tubercle corpuscles" vary somewhat in size and form. Many are 1–2,000th of an inch or less in their longest diameter. They become indistinct if immersed in glycerine, and are rendered very transparent by potash, and also by acetic acid. It is only during the early period of the development of the tubercle corpuscle that anything that could be fairly termed a "nucleus," or "nuclei," can be displayed by the action of acetic acid. The tubercle corpuscles are not free nuclei. Amongst these special corpuscles by which tubercle is distinguished from other pathological elementary parts, and especially in the case of old tubercles, much granular matter, consisting partly of calcareous and partly of fatty matter and oil globules, is to be seen—and in most specimens pus-corpuscles are also present. The student must be prepared to find many anatomical elements as well as the so-called tubercle-corpuscles; and the number of pus corpuscles, and the proportion of granular matter and oil globules will vary according to the age of the tubercle and the amount of degenerative change it has undergone. Occasionally in the caseous material that results from the disintegration of tubercle, minute but well formed crystals of cholesterine may be discerned, but almost invariably if a little caseous tubercle be treated with warm alcohol crystals of cholesterine may be obtained by concentrating the filtered alcoholic solution.

Little, however, is to be learned concerning the growth and multiplication of tubercle corpuscles by examination with a quarter of an inch object-glass. And it is a mistake to suppose that in order to form an accurate conception of the development and growth of this, or indeed any morbid bioplasm, a great number of cases must be examined. On the contrary, the observer should obtain a few good specimens of the diseased structures he desires to examine and study them thoroughly. Portions should be prepared according to different methods, and the specimens carefully compared with one another.

Many careful investigations have been made upon tubercles artificially induced in the lower animals, but such tubercles differ in some general points, and also in some microscopic characters from the tubercles developed in the course of disease in the human subject. Upon the whole, therefore, I think our conclusions should be based principally upon the results of observations upon human tubercle.

In tubercular disease of the pia-mater of the human subject beautiful specimens of tubercle, in every stage of development, may be obtained; and the constituent textures of the pia-mater render it a better tissue than the lung for studying the minute pathological changes occurring in tubercle.

Most observers who have studied tubercle have employed comparatively low magnifying powers, and their drawings, instead of help-

ing us to understand how a tubercle corpuscle lives and grows, represent collections of thousands of little bodies like one another, from which little is to be learnt save that a young "tubercle" certainly consists mainly of the corpuscles in question.

To form an accurate notion of the shape, general aspect, mode of increase of the corpuscles, and their relation to adjacent structures, objectives, magnifying at least five hundred diameters, are required, and the methods of investigation above referred to will not be found so advantageous as the plan I have recommended in page 61, which is admirably suited for the investigation of this morbid structure. I recommend the student to inject a portion of a child's tubercular lung first with the carmine, and then with the Prussian blue fluid, page 105. After small pieces have been soaked for a week or two at least in acid glycerine, the precautions given in the upper part of page 101, being carefully observed, very thin sections may be made and preserved permanently in glycerine containing two or three drops of acetic acid to the ounce. Specimens thus prepared may, by remounting and carefully applied pressure, be frayed out so as to show many points not usually represented in published drawings. They are always to be examined in the strongest Price's glycerine, containing about five drops of glacial acetic acid to the ounce. The examination may be conducted with a twelfth and afterwards with the aid of a sixteenth or a twenty-fifth. Many of the "tubercle corpuscles" will be found to be neither oval nor circular —and here and there a particle will be discovered which is undergoing division or giving off diverticula. In fact, by the process of investigation I have recommended particles of bioplasm may be preserved and mounted permanently, exhibiting the form they had when taken from the living body. Appearances seldom seen, though constantly occurring during the process of multiplication of particles of bioplasm, are not only in this way made very distinct, but the specimens preserved permanently. I have elsewhere figured a beautiful specimen of cartilage bioplasm in process of division. See "Protoplasm," 3rd Edition, pl. VI.

301. Of a "Tubercle."—A "tubercle" is a body of complex structure, and almost invariably consists, not of special and peculiar anatomical elements only, but of the more or less changed tissue elements in the seat of the development of the tubercle, as well as of bioplasts and products characteristic of inflammatory change.

The many different constituents of a tubercle will vary in proportion under different circumstances, and at different periods of the growth of the tubercle. If a tubercle grows very quickly, it will differ in many points of structural detail from one which has been very slowly developed. A young growing tubercle is surely so unlike the yellow cheesy matter resulting from the disintegration and decay or obsolescence of tubercle that we cannot but wonder that such different things

should be known by the same name. The difference is not less than that which marks off the acutely-growing cancer from the lifeless pasty granular matter which sometimes results when a portion of a cancerous tumour has died and undergone disintegration and other changes.

It is obvious that tubercle cannot be derived from caseous matter as many have insisted is the case, for tubercle is alive and capable of growing and multiplying, while caseous matter is dead and is incapable of growth. It cannot propagate itself, and is a perfectly passive substance. There is no analogy between the growing tubercle and the caseous matter. The latter is but a product resulting from the death and decay of the former. But it is by no means the constant result of such changes. Nay, caseous matter is neither constantly associated with the production of tubercle, nor is it peculiar or confined to this form of morbid growth. Caseation may occur independently of tubercle, and the latter may run its course without giving rise to the formation of a particle of cheesy material.

It has often been said that tubercle is due to inflammatory change, but so far from this being the case, the development of the specific form of bioplasm known as tubercle is not usually associated with any form of inflammation. Nay, inflammatory action would interfere with, if it did not entirely prevent the development of true tubercle. Inflammation may succeed or precede the origin of tubercle, and the inflammatory process may be caused to commence by tubercle, but the development of tubercle is a morbid phenomenon quite distinct from and independent of inflammation. As is well known, inflammatory changes may affect cells, vessels, and other textures, and may proceed for any length of time without the development of a single tubercle bioplast while the latter may originate in the same tissues without any inflammation whatever. Indeed in tubercular and inflammatory change there is little in common. Tubercle, then, cannot be, as has been often asserted, "an inflammatory product." Neither is tubercle a mere overgrowth of elements of normal texture, nor can it be properly regarded as adenoid tissue, for the latter may be developed under conditions which are widely separated from those associated with the tubercular state.

A tubercle is not an unusual development of lymphatic tissue, as is maintained by many. As far as I can ascertain, there is no analogy between tubercle-corpuscles and lymph-corpuscles. Lymphatics are less numerous in the lung tissue and in the pia-mater than in many other textures, which are very rarely invaded by tubercle. Nor do we discover in those cases in which tubercles are very abundant in the lungs that they are more numerous or larger in those situations where lymphatics abound, than in those parts of the pulmonary organs in which very few are to be discovered. In fact the lymphatic doctrine of tubercle

rests for the most part upon conjectural facts, observations, and discoveries. Much of our modern pathology, it seems to me, is founded upon the vague statements of reputed observers, who, while professing to state what they have seen, are, in point of fact, merely giving their own interpretation of what has been seen and described by other observers. This lymphatic doctrine is but a modified form of the old idea successfully controverted by Matthew Baillie that tubercles were developed from the lymphatic glands.

Vessels never grow in connection with tubercle, but on the contrary many are completely destroyed during the process of tuberculization. Capillaries, and indeed small arteries and veins, may be found embedded in a mass of tubercle, but they are either the vessels belonging to the original tissue, or vessels which have been developed in lymph in which tubercle was subsequently deposited. Many years since, Quekett made some beautiful injections of tubercular lung, in which the walls of air-cells could be traced in the substance of the tubercle by the injected vessels. In this respect tubercle differs absolutely from those morbid growths, which may be regarded as modified normal tissue (many forms of tumours and cancers), and perhaps more nearly approaches such morbid bioplasm as pus.

The term "infiltration" is inaccurate, and does not convey a correct idea of the process which takes place in the formation of a tubercle. We may speak of a tissue being infiltrated with a material already formed and suspended in the fluid which traverses the tissue, or with a material precipitated from solution in its interstices, as lime salts in the organic matrix of bone. But to speak of a texture being infiltrated with pus, tubercle, or cancer, is, as it seems to me, incorrect, as these are all forms of living bioplasm, and are *active*. They *grow* instead of being deposited, and we might as well speak of the earth being infiltrated with the roots of a tree as of the lung or other organ being infiltrated with tubercle. "Grey infiltration," "yellow infiltration," "gelatinous infiltration," "caseous infiltration," are all highly objectionable phrases, and if used are certain to mislead by conveying an incorrect idea of the actual phenomena.

There are, as is well known, different kinds of pus and cancer bioplasm, and there are forms of tubercle bioplasm which differ from one another, at least in as great a degree as certain forms of pus and of cancer differ from other forms. But not only may it be fairly held that there are different kinds of tubercle as of pus and cancer, but, as in the case of certain forms of cancer, the course which the tubercle-corpuscle takes may be very different. This may be admitted, but if we go so far as to assert that the tubercle-corpuscle may form fibroid or cartilaginous tissue, I think we may be fairly accused of begging the question at issue. Are we sure that these things have been formed by

an actual tubercle-corpuscle—by the "tubercle-bioplast?" In tubercle of the pia-mater, it must be borne in mind, we never do find abnormal textures like those supposed to result in certain instances from tubercular change in the lung. It is therefore at least open to question whether these fibroid and cartilage-like textures found in the lung in certain cases do not originate from other than the special tubercular bioplasm which constitutes the principal morphological constituent of a true tubercle.

302. The Tubercle Bioplast.—Tubercle, like pus, lymph, and white blood corpuscles, and the contagion particles considered in p. 320, while in an active state, is living, and like every other form of bioplasm, has been derived from living matter. Although the precise nature of the phenomena occurring just antecedent to the origin of the tubercle bioplast, and to which its production is to be referred, is unknown, it is quite certain, 1, *that all tubercle springs from living matter;* and, 2, *that like pus, it may originate in bioplasm* that has apparently existed up to that time in a normal state. That the tubercle-corpuscle is living growing bioplasm is proved most pointedly in the case of tubercular disease of the pia-mater. There is no caseous matter to be seen in many cases of this affection; nor is there débris, or anything having the character often assigned to tubercular material. Multitudes of minute bioplasts grow very actively upon the vessels, particularly in the external coat of the small arteries in this disease, and in many instances form minute collections, the several particles constituting which may all have descended from one.

The tubercle bioplast is a minute particle of living matter, which is of much firmer consistence, and which grows much more slowly than a pus corpuscle. It is certain that pus may be formed from normal bioplasm in less than fifty hours, but it is doubtful whether a living particle of tubercle could be developed in as many days. As a general rule, the several stages of its development unquestionably require for their completion a much longer period of time; and it is probable that many successive series of bioplasm particles are developed before the remarkable properties characteristic of the tubercle-bioplast are acquired or developed to their full extent. In this process it is probable the new centres (nuclei) successively developed in the bioplasm play an essential part. The new powers are probably acquired while the bioplasm exists as the nucleus of the bioplasm particle already formed. This nucleus arises or makes its appearance in the central part of the particle. Sometimes several new centres or nuclei originate in a single particle of bioplasm. These at first slowly grow, but as the bioplasm outside gradually changes, the nucleus becomes larger, and at last assumes the characters of a bioplast, in which new centres are in their turn developed.

The tubercle bioplast, as ordinarily observed, is irregular in form, and is often more or less angular. It may be oval, but is never perfectly spherical. Sometimes the bioplasm grows and divides and subdivides so as to form compound masses which are really collections of tubercle-corpuscles. This is probably the nature of the bodies which have been termed "giant-cells," first described by Recklinghausen. The mode of growth and multiplication of a tubercle-bioplast has been already referred to in the last page. It is seldom that the changes there referred to can be demonstrated in the lung, but upon the vessels in tubercular disease of the pia-mater I have been able to demonstrate the dividing bioplasts very distinctly, and in several instances. See fig. 3, pl. XLVI.

Tubercle bioplasts having once been formed, may spread—1. By being detached from the general mass moving away from the spot where they were developed, and then growing and multiplying—and, 2. By gaining access to the lymph or blood, and being carried in one or other circulating stream to distant parts of the body.

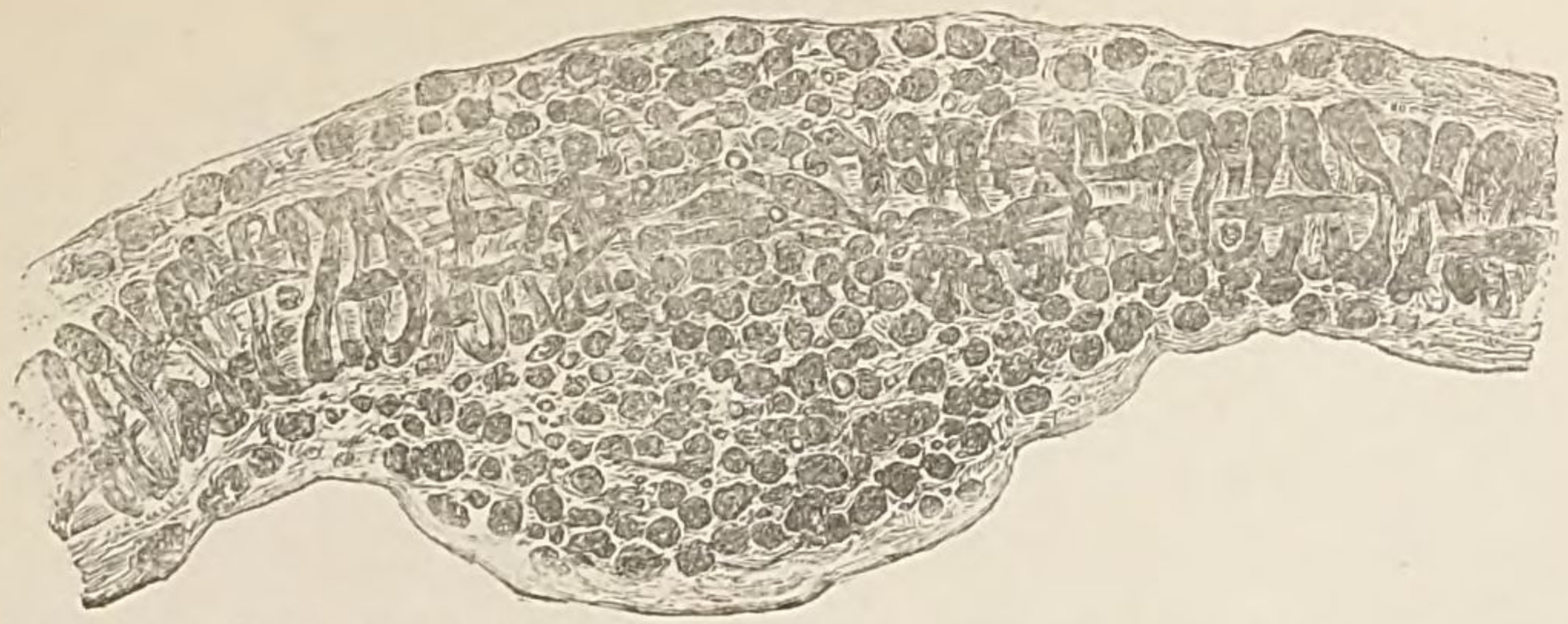
The matter around the Tubercle Bioplast.—It has been already remarked that the bioplasm of the tubercle-corpuscle undergoes change upon its surface, and becomes converted into a passive material, which differs in character in different cases—sometimes appearing as mere granular débris, sometimes as a delicate, or moderately firm tissue. Indeed a greater or less proportion of a delicate fibrous material may be almost invariably found. This has been described by some as a reticulated structure, in the meshes of which the tubercle corpuscles are found, but I doubt whether it ought to be regarded as a special structure belonging to tubercle. I incline to the opinion that it is an imperfectly developed formed material, perhaps analogous to the fibrinous matter which is found in connection with white blood, and lymph corpuscles, and many other forms of bioplasm. The blood clot is reticulated, but it can hardly be correctly spoken of as a tissue.

This formation of a fibroid material then is not peculiar to tubercle bioplasm, but lymph corpuscles and white blood corpuscles under certain circumstances will produce a form of fibrillated tissue. The more slowly the tubercle bioplast grows and multiplies, the firmer and more condensed will be the formed material that results; and it seems probable that the very bioplasts which, under certain conditions, simply grow and multiply, and lead to the disintegration and breaking down of adjacent textures, under other conditions may multiply much more slowly, and may give rise to a passive structure. This may result in puckering, contraction, and condensation of tissue, but such a tissue does not soften or determine softening and disintegration.

303. Of Giant Cells.—The "giant cells" are not *cells*, but as regards their formation may be compared with the so called *myeloid cells*

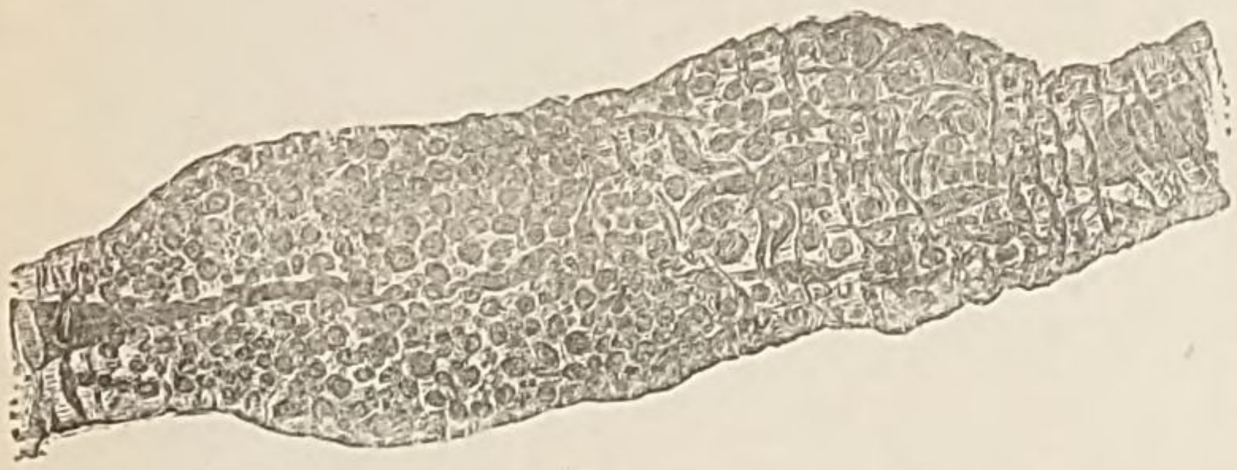
TUBERCLE BIOPLASM. PLATE XLVI.

Fig. 1.



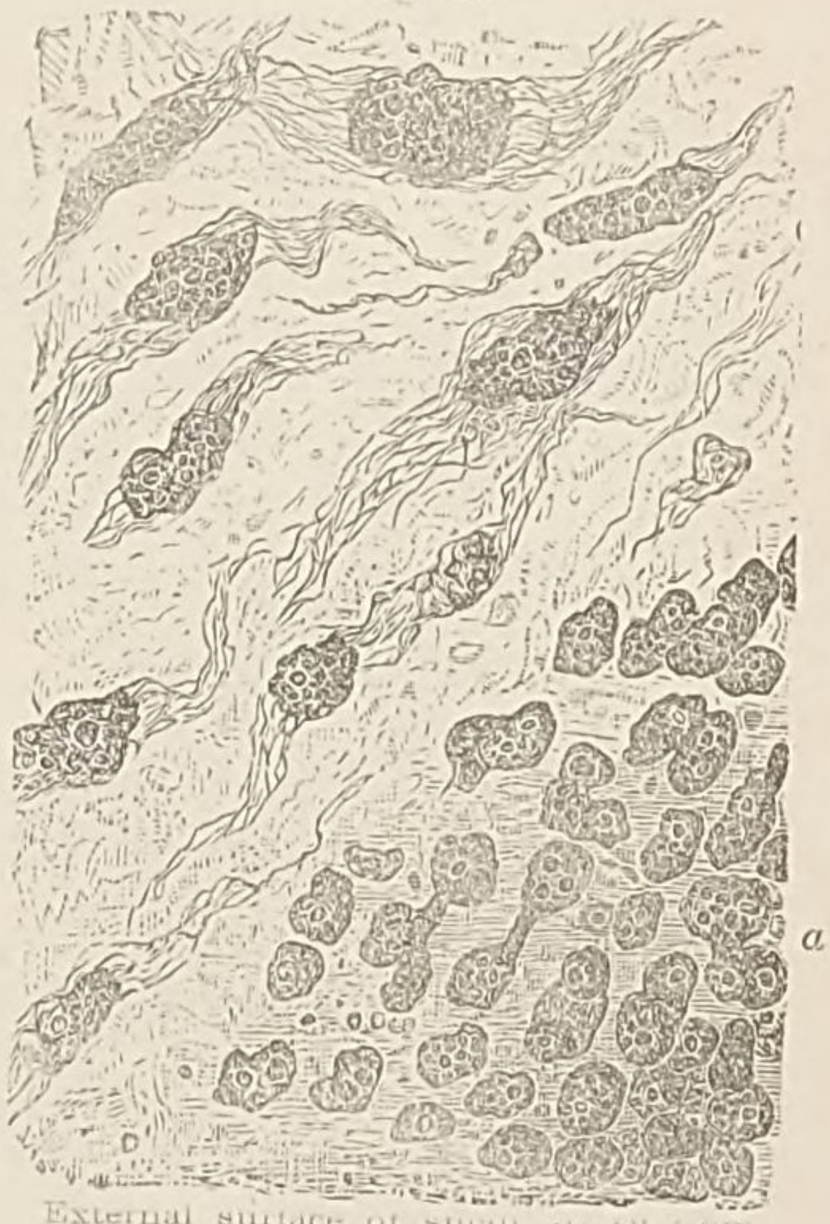
Tubercle bioplasts in the outer coat of a small artery. Tubercular inflammation of pia-mater. Death three weeks after commencement of the symptoms. × 215. p. 333.

Fig. 2.



Artery from tubercular disease of pia-mater, showing alterations in the external coat of the vessel and obliteration of the cavity. × 130. p. 333.

Fig. 3.



a

External surface of small artery from tubercular disease of pia mater, showing the enlarged bioplasts of the connective tissue, and at *a* tubercle corpuscles growing and multiplying rapidly × 700. p 330.

Fig. 4.

Tubercle corpuscles from a tubercle in the lung. × 215 p. 329.

Fig. 5.

Tubercle corpuscles from the interior of the eye of a child aged eight. Removed by Mr. Hulme. × 215. p. 329.

Fig. 6.

Increase of bioplasts of the capillaries of tubercular lung. Below *a* numerous tubercle bioplasts are seen just outside the capillaries, and in some places the vessels are distended by the growth and multiplication of the particles inside. p 334.

[To face page 330

of bone. These bodies are not found either in veins or in lymphatics, as has been suggested, nor are these or any other "cells" produced by the fusion of the epithelial lining of an alveolus of the lung tissue "into one protoplasmic lump," as has been affirmed by Dr. Klein.*

The so-called "giant cells" are sometimes very large, according to Virchow, as much as the 1-300th of an inch in diameter—but it seems to me doubtful whether these or the so-called "myeloid cells" in bone ought to be termed cells. The bodies in question result from the growth and division and subdivision of bioplasts in a moderately firm formed material, which is continuous throughout the mass. I have seen many excellent specimens of tubercle which were quite destitute of "giant cells;" and I cannot assent to the view that they are in any way characteristic of tubercle, or when present, play a necessary or essential part in the pathological changes which distinguish tubercular disease.

304. Probable origin of Tubercle.— I have conclusively proved that every kind of tubercle, at least at an early period of its growth, consists of bioplasm; that every form of bioplasm comes from pre-existing bioplasm may be accepted as a fact not very likely, I venture to think, to be upset by facts discovered by future investigation. The proof that the morbid bioplasm of tubercle comes from bioplasm that did not possess the powers that characterise tubercle is at least as distinct as the proof that pus may be derived from the normal bioplasm say

* "Reports of the Medical Officer of Health," new series, 1874, p. 92. Such unfortunate descriptions introduce new confusion into the already sufficiently confused new Pathology. I would only venture to remark here, 1, that epithelial cells never, under any circumstances, become fused together, neither do they form such a body as a "giant cell;" and 2, that protoplasmic "lumps" have been probably suggested by the belief in the hypothetical bathybius, which exists in the imagination of a very few observers. The expression is highly objectionable, for no lumps of any kind, nor any bodies whatever that can be correctly spoken of as "protoplasmic" are formed in the manner stated, or found under the circumstances named. The reader may form a notion of the sort of pathological doctrines we are expected to accept in England if he will study the following paragraph :—

The changes in alveolar cavities containing giant cells are said to be as follows :— "The cylinders of multinuclear protoplasm grow and divide into a number of giant cells, which gradually become converted into a tissue to a certain extent resembling adenoid tissue, but differing from it in many respects. Thus the giant cells give origin to a more or less regular network of nucleated cells, which *consisting at first of granular substance*, soon assumes the appearance of a more or less distinct fibrillar substance; in their meshes lie only a limited number of lymphoid cells. This tissue spreads very rapidly, and finally undergoes, from the centre outwards, a fibrous degeneration, which becomes the seat of cheesy deposits."—"Reports of the Medical Officer of the Privy Council," new series, No. III, p. 89.

Most of the statements in the above paragraph rest, as it seems to me, upon a very slender foundation of fact. It is much to be regretted—indeed I think it hardly right that such views should be authoritatively promulgated by the Medical Officer of the Privy Council in a Parliamentary Blue Book.

of epithelium or of connective tissue, as well as form other forms of tissue bioplasm.

I do not propose to discuss the modern doctrine of the origin of tubercle from endothelium, because I do not agree with the views upon which this doctrine rests. Although I have seen what has been termed "lymphatic endothelium," I cannot look upon serous cavities as colossal lymph sacs. Nor can I regard tubercle as modified endothelial cells, because it is the bioplasm only of the so-called endothelial cells from which tubercle corpuscles could possibly be derived. Tubercle corpuscles, at least during the early and active period of their growth, cannot be properly termed "cells;" and although the outer part of the tubercle corpuscle may become somwhat hardened, it does not possess characters which justify the application to it of the term cell-wall.

As regards the question of what particular normal bioplasm is the seat of origin of the tubercle corpuscle, there is room for speculation—and although in some instances we may seem to have evidence of its derivation from a special form of bioplasm, further investigation will probably lead us to question the correctness of such a conclusion, and may rather favour the idea that tubercle, like pus, may originate in more than one form of bioplasm. I have preparations which support the view that tubercle sometimes originates in the bioplasm of the capillary wall; others, again, seem to show that it starts from connective tissue corpuscles (pl. XLVI, fig. 3). Lymph corpuscles are regarded by many recent investigators as the progenitors of tubercle, and even epithelial cells are by some supposed to be the exact seat of its origin. Certain minute particles of bioplasm, which, escaping from the blood, traverse the capillary wall, and grow and multiply outside the vessel, seem, in some instances, to be the sources of tubercle. I could advance some strong arguments in favour of the view that tubercle bioplasts may exist as such minute particles of living matter, and may be carried suspended in blood lymph or chyle to parts of the body at a distance from the seat of their origin.

There is no doubt that in some cases the earliest changes which take place in the development of tubercle occur in connection with the bioplasm of certain capillary vessels. It is in tubercular disease of the pia-mater that early changes may be very clearly demonstrated, as I showed in the course of some observations made many years ago. In some specimens, the so-called nuclei, that is, the oval bioplasts of the capillaries are seen to be much larger than in the normal state, and here and there an enlarged bioplast may be found which is actually undergoing division.

In ordinary inflammations and in fevers (pl. XXXVII, figs. 5 and 6, page 264), the bioplasts of the capillary vessels also increase in size, and the process may result in the formation of pus. Now the enlarge-

ment occurring in fever and in inflammation could not be distinguished from what takes place in the development of tubercle, but the latter change proceeds much more slowly than the former one.

I have assured myself that in some instances very early changes in the development of tubercle occur in the bioplasts of the connective tissue forming the external coat of very small arteries. These bodies may be enlarged over a very limited area, sometimes only from three to six being involved. After a time, the enlarged bioplasts divide and subdivide, and thus results a small collection of particles (pl. XLVI, figs. 1, 6).

Again, in other specimens the earliest changes appear to affect the bioplasts very near to the muscular coat of the small artery in the first instance. But the facts might be explained on the supposition that the morbid bioplasts did not result from the enlargement of connective tissue corpuscles at all, but depended upon the growth of minute particles which had made their way out of the lymphatics, or from the blood, traversing the arterial wall in the intervals between the muscular fibre-cells of the vessel.

Important changes take place in the interior of the little artery, the external coat of which is the seat of development if not of the origin of tubercle. The flattened spindle-shaped epithelium-like particles which line the vessel are soon detached, and collect in the tube the calibre of which has already been greatly reduced. The vessel is in this way completely obstructed, and the branches beyond the point of obstruction soon begin to waste and to undergo degeneration. The muscular fibre-cells which surround the artery are often separated from one another, and exhibit the same appearance which is often seen in vessels which have been over-distended in artificial injection. As regards the structure of the coats of the very small arteries, there is nothing to be urged against the view that minute particles of bioplasm may make their way from the blood, and passing in the intervals between the muscular fibre-cells, reach the outside of the vessel, and grow and multiply in the areolar coat, thus forming bodies which at a certain period of their growth could hardly be distinguished from the connective corpuscles themselves.

The view now most in favour is that in which the bodies, formerly regarded as connective tissue corpuscles, are considered to be cells of endothelium belonging to lymphatic spaces on the external surface of the arteries. I cannot, however, concur in this doctrine, because I have in many instances traced the gradual enlargement of the bioplasm of the connective tissue corpuscles, and have seen corpuscles exhibiting various gradations in size in a very small space. In ordinary inflammation, the changes may be studied with facility—particularly in the frog, and one may feel sure that it is the bioplasm of connective tissue—not of endothelium, which undergoes the enlargement.

From several specimens I have seen, I am convinced that tubercle-corpuscles may originate in the bioplasm of the capillaries of the peritoneum, pleura, and other serous membranes, and in the bioplasm which exists in the walls of the capillaries of the lung and of the areolar tissue between the lobules of the lung, in the bioplasm of the capillaries of the pia-mater, liver, kidneys, and other organs.

The evidence of tubercle being universally lymphoid in its origin is inconclusive, while there is no justification for the opinion that it is invariably derived from lymph-corpuscles, and that tubercular disease always depends upon morbid changes connected with the lymphatic system. It used to be supposed that pus could originate only in connective tissue corpuscles, or in these and in epithelial cells only. But investigation convinced me as long ago as 1860, that pus might result from any bioplasm of the body. There are, however, certain forms of bioplasm which very rarely give rise to pus, while close by these may be some which very readily take upon themselves increased action. Still pus may originate in any. I think that as time goes on we shall be led to a similar conclusion as regards tubercle, and that instead of invariably originating in one form of bioplasm, it will be conclusively demonstrated that many may, under certain circumstances, give origin to it.

Now, at least in certain forms of tubercular disease, similar living bioplasts to those enlarged in inflammation are affected—but the increase in size takes place much more slowly. We have no facts to justify the conclusion that the development of pus or contagious virus, or tubercle, or cancer, from normal bioplasm, is determined by the state of the blood, while many arguments might be advanced in favour of the doctrine that tubercle and cancer are, at least in some instances, due to developmental phenomena, while pus may arise in any bioplasm whatever, and at any period of life: virus being produced under certain peculiar and often persistent and exceptional conditions of life. There is much to be urged in favour of the view that the formation of tubercle is not due to the circumstance that normal bioplasm receives some peculiar pabulum, or lives and grows under certain peculiar conditions. The origin of tubercle cannot, like that of pus, be explained simply by the increased access of pabulum to the bioplasm; and it might be urged that a review of the facts rather inclines us to believe that in cases in which tubercle is formed there existed from the first some inherited peculiarity in the bioplasm which gave rise to it. All persons, it might be said, would not develop tubercle, even if placed under circumstances the most favourable for its development. But I confess such arguments seem to be entirely subverted by the fact, that wild animals, when shut up in confined rooms, frequently do fall victims to tubercular disease. Monkeys are particularly prone to this morbid change; and it is practically certain that in tubercular

monkeys the normal bioplasm must have developed the tubercular characteristics, and that the tubercle corpuscle must, at least in this case, have been produced direct from normal bioplasm. I think, therefore, we must conclude that although in the majority of instances there has been from the first a tendency, probably inherent, on the part of bioplasm, to develop tubercle, this may, in certain instances, originate from the normal bioplasm of a perfectly healthy organism brought up under bad hygienic conditions. On the other hand, I regard it as almost certain, that if persons possessing bioplasm prone to assume the tubercular state are placed under such circumstances as are known to retard or interfere with the transformation, they may escape and live to be very old without any formation of any true tubercle-corpuscles at any period of life. Nay, there is good reason to think that in certain instances in which tubercle has actually been developed and the living particles of morbid bioplasm have even grown and multiplied, to a certain extent, the further progress of the disease—or in other words, the further growth and multiplication of tubercle-bioplasts—has been arrested; and this in consequence of the organism having been placed under circumstances the very opposite of those which are known to favour tubercular change.

Lastly, I will remark that in whatever form of bioplasm the tubercle bioplast may originate, its production is invariably associated with more rapid change than characterises the normal state. Too much bioplasm is produced—too little undergoes change into formed material. Though quick as compared with healthy changes, the change is slow indeed contrasted with the rapid changes taking place when bioplasm develops pus corpuscles. Tubercle bioplasm, besides growing and multiplying, degenerates more slowly than pus. It is almost certain that a diet rich in fatty matter, retards the growth and multiplication of tubercle corpuscles, probably because elements of the fatty matter, in some way combining with nitrogenous constituents which would otherwise be appropriated by the tubercle corpuscles, and contribute to their growth and multiplication, assist to form more evanescent forms, resulting in the production of chemical compounds which can be readily eliminated from the organism.

305. Change and Degeneration of Tubercle—Caseation—Fibroid Change.—Tubercle corpuscles, like all other things, having lived for a certain time, die, and become changed. They may lose water and be broken down into granules, which contain albuminous matters, and fatty matters which are rich in cholesterine and earthy salts. Or, as happens only too seldom, the products of the disintegration may be entirely absorbed, and the tissue invaded by the tubercle may return to its normal state.

One of the commonest changes which tubercle undergoes is "casea-

tion," in which a soft material, consisting of granular matter, composed of earthy phosphate, and oil granules and globules with débris of tissue elements, and not uncommonly small crystals of cholesterine results. This cheesy material used to be considered peculiar to tubercle. Indeed tubercle was regarded as cheesy matter until some years ago, when Virchow pointed out that pus and other inflammatory products, as well as cancer, might undergo the process of caseation. Tubercle, instead of undergoing change into caseous substance, may gradually be succeeded by a fibroid material, which, save condensation, seems to undergo little change. This fibroid change is progressive, and may last for many years, gradually involving a considerable extent of pulmonary tissue. In some cases, as tubercle undergoes obsolescence, a different kind of formed material, more like cartilage than any fibroid tissue, results.

According to the views entertained by many the tubercle corpuscle itself may grow and form fibroid or cartilage-like material, or die and degenerate, caseous matter being among the materials resulting. Important alterations are no doubt caused in the adjacent apparently healthy lung tissue by the growth and subsequent changes in the tubercle. The capillary vessels in the neighbourhood become obstructed by the formation of coagula within them, and with their contents at last undergo disintegration, and are removed. The elastic tissue and other anatomical elements entering into the formation of the lung are changed, and at last disappear.

306. Views concerning the Nature of Tubercle.—There has always been very great difference of opinion as regards the nature of tubercle, and at this time, although our investigations may be carried to a degree of minuteness not considered to be attainable many years ago, pathologists are as much divided as ever. Some regard tubercle as matter resulting from the decay or degeneration of some pre-existing material. Tubercle has been considered to be mere débris, and it has been stated that it is soluble in water. Some have looked upon it as an excretion, and by many it has been included among the exudations. The tubercle corpuscles, it was thought, were aggregations of minute lifeless granules precipitated from a fluid poured out from the blood. "Inflammation," the reputed source of so many pathological products, has been set down as the cause of the development of tubercle, but, as already remarked, the inflammatory process is as distinct from tuberculization as is the pus corpuscle from the tubercle bioplast. There are many forms of tubercle and many forms of pus, but every form of the one is distinguished in the most marked way from every form of the other. Although different kinds of morbid bioplasm assume the form of very minute living particles that could not be distinguished by microscopical examination, it would be idle to argue that therefore pus, tubercle, the virus of various contagious fevers, the poison of syphilis, cancer, and some others, are

allied and all related to pus, which results from the process termed "inflammation." This "inflammation" leads to the production of the pus class of bioplasm only, and all the other forms of morbid bioplasm named, originate and acquire their specific properties independently of any inflammatory process.

Of course "irritation" has been confidently asserted to be the efficient cause of the production of tubercle. But "irritation" no more accounts for the formation of tubercle than it does for that of pus, or cancer, or the virus of a contagious disease. In fact, "irritation" is a process that has not been defined. The word is in constant use, but no one who uses it considers it necessary to define what he means by it. More recently we have been assured that tubercle results from lymphatic proliferation produced by "irritation," an assertion which rests upon several conjectures, all of which are unproved, and some of which are unprovable. That the bioplasm of tubercle grows faster than the normal bioplasm in which it originates is probable, while it is certain that it grows and appropriates nutrient matter more slowly than pus does, but such a fact establishes no connection whatever between pus and tubercle, or between inflammation and tuberculization. That it is impossible to distinguish the tubercle corpuscle from many forms of inflammatory bioplasm may be readily admitted, but it is equally impossible to distinguish other forms of bioplasm from one another by their microscopical character, or by chemical tests. Tubercle has been referred, like many other pathological growths, to chronic inflammation and to local inflammation set up by "irritation," but its life history is totally distinct from that of any inflammatory product known. If the observer carefully examines a young tubercle formed upon a serous membrane, or in connection with the vessels of the pia-mater, he will be able to prove conclusively that the little particles of which the growing tubercle consists, are composed of living bioplasm. Each one may grow, divide, and subdivide, and no doubt minute tubercle corpuscles in a living state may pass from the seat of their production through the capillary walls, or into lymphatic vessels, and be carried to distant parts where they may become fixed and germinate and grow and multiply till more tubercles are produced.

Question of Contagiousness of Tubercle.—Much has been written upon the question of the contagiousness of tubercle, and many experiments have been carried out in the hope of obtaining definite conclusions. The inoculation experiments upon animals are not conclusive, because other substances besides tubercle give rise to similar phenomena, while the cases in which one person is supposed to have caught the disease from another are devoid of anything approaching to proof. Upon one hand, it is very improbable that lifeless caseous matter and the substances generally known as tubercle, should generate the specific disease, upon

the other it is most unlikely that growing living tubercle bioplasm particles should be detached, and become mixed with sputum, although it is almost certain that if these were properly inoculated in a favourable situation, they would grow and multiply and produce a tubercle like that from which they were derived.

307. Of the Three kinds of Morbid Bioplasm—Pus, Contagium, Tubercle.—In concluding this chapter upon three kinds of morbid bioplasm, which I believe have all been derived from the normal bioplasm of the body—by direct descent, I will remark that in all healthy and morbid actions in living beings, the agents directly concerned are minute particles of bioplasm. In all vital phenomena, in all organisms, such particles of living bioplasm play an essential part. They are composed of clear transparent material, and when examined under a magnifying power, of 5,000 linear, appear to be perfectly devoid of structure. The living matter or bioplasm of which these particles, possessing such diverse powers consists, exhibits the same appearance. Nay, it is not even possible to distinguish by microscopical examination, or with the aid of chemical tests, a healthy from a morbid particle. Nor would the most skilled observer be able to determine by physical examination whether a particular normal particle of bioplasm had belonged to a plant, to an animal, or to man.

The particle of contagium can be distinguished from normal bioplasm only by the fact of its giving rise to a disease in the organism into which it gains an entrance, in all respects like that affecting the organism from which it was derived,—but then the embryonic bioplasm of a dog is to be distinguished from that of a man by the fact of its developing into a dog only.

Each form of normal bioplasm has its peculiarities as regards the pabulum on which it feeds, the rate of its growth, the temperature most favourable to its development, and the external conditions necessary. And similarly, every different form of morbid bioplasm has its individual characteristics. Pus grows faster than tubercle, and the latter probably faster than many forms of cancer. Pus may be formed under very simple conditions, which are known—tubercle under more complex conditions, concerning which we have much to learn, and cancer under circumstances more complex still. Of pus, there are general and specific kinds; of tubercle and of cancer there are also different forms. The causes of the differences in the case of pus are not referable to the developmental period of life, while in the case of cancer they are probably invariably so. Tubercle may or may not be due to phenomena occurring while the early changes connected with development are proceeding.

How, it will be asked, can a little speck of living matter be the efficient cause of small-pox, or measles, or fever, or ophthalmia, or

phthisis, &c. ? There is nothing distinctive about it, neither can I see any reason whatever for supposing that anything distinctive will ever be discovered. But at least it must be admitted that until we have discovered how to distinguish a living particle of a dog germ from one of a man germ, there will be little prospect of our being able to determine the points by which the small-pox, or scarlatina germ, may with certainty be distinguished from the measles, or typhus, or other fever-germ ; or the tubercle germ, or pus germ, from any one of them. In the present state of our knowledge, the only answer to the question, what causes the remarkable individual characteristics of all these different particles, seems to be that the differences in properties and powers manifested by the several particles respectively, are not due to any *material* properties but to peculiarities of *vital power*, communicated by pre-existing living particles.

CHAPTER XVI.

ON URINE, URINARY DEPOSITS, AND CALCULI.—*Collecting Urine for Microscopical Examination.—On Examining Urinary Deposits in the Microscope.—Magnifying Powers.—Chemical Examination of Urinary Deposits.*—EXTRANEOUS SUBSTANCES MET WITH IN URINE.—OF URINARY DEPOSITS.—*Mucus.—Vibriones.—Torulæ.—Penicillium Glaucum.—Sugar Fungus.—Epithelium.—Spermatozoa.—Casts of the Uriniferous Tubes; of Medium Diameter; of Considerable Diameter; of Small Diameter.—Fat Cells.—Conditions in which Fatty Matter occurs in Urine.—Pus.—Earthy Phosphates.—Urates. Uric Acid.—Oxalate of Lime.—Dumb-bells.—Triple Phosphate.—Cystine.—Carbonate of Lime.—Blood Corpuscles.—Large Organic Globules.—Small Organic Globules.*—URINARY CALCULI.—*Formation of Calculi.*—ON THE PRESERVATION OF URINARY DEPOSITS; *in the Dry Way; in Canada Balsam; in Aqueous Solutions.*

IN this work, I shall only give a very short summary of the microscopical characters of some of the principal urinary deposits, and describe the mode of collecting them and the processes adapted for their preservation. The microscopical and chemical characters of the urinary constituents in health and disease have been fully described in another work, to which the reader is referred for more complete information.* Drawings of those deposits which most frequently come under the notice of the practitioner will be found in the plates.

The microscopical examination of the urine has of late years become a subject of such great importance, and the advantages derived from it are so generally admitted, that it is unnecessary to dwell upon the matter. Within the last fifteen or twenty years, the investigation of urinary deposits has been so much simplified by the use of the microscope in conjunction with chemical analysis, that the nature of the greater number of deposits has been correctly ascertained. The investigations of Dr. Prout, followed by those of Drs. Golding Bird, Jones, Christison, Owen Rees, Johnson, Roberts, and many others, have shown the importance of the examination of the urine, and the advantages derived from it in the diagnosis and treatment of urinary diseases.

* "Kidney Diseases, Urinary Deposits, and Calculous Disorders; their Nature and Treatment." Third edition, 1869.

By frequent examination of different specimens of urine, and reference to the drawings in this book, the student may soon become familiar with many of the deposits he is likely to meet with. At first, however, he must be prepared to encounter serious difficulties, some of which it is desirable to point out at once. In some specimens of urine which he examines, he will perhaps be surprised to find no deposit whatever, whilst in examining others, the whole field of the microscope appears to be occupied by substances of various shapes and colours, the nature of which it is not easy to ascertain by reference to works on the subject. Many of the bodies whose presence occasions this difficulty, have obtained entrance into the urine accidentally, and these are often mistaken by the student for urinary deposits. Portions of hair have been mistaken for casts of the renal tubes, starch granules for cells; and other substances of extraneous origin, such as small portions ot woody fibre, pieces of feathers, wool, cotton, &c., not unfrequently take the form of certain urinary deposits, and to a certain extent resemble the drawings of them in their general appearance, so that the student is misled in his inferences, and puzzled at the very commencement of investigation.

The principle followed in this book, namely, that of supposing the student actually engaged in working at that part of the subject under discussion, has been adhered to as far as practicable in this chapter.

308. Collecting Urine for Microscopical Examination. — Urine, which is to be submitted to examination, should be collected in considerable quantity (not less than four ounces), in order to obtain sufficient of the deposit for microscopical investigation. In many instances the amount of sediment, even from a pint of urine, is so small that, without great care in collecting, it may be altogether passed over. The bulk of deposit from a measured quantity of urine should always be roughly noted. The space occupied by it may be compared with the total bulk of the liquid, and we may say the deposit occupies a fifth, a fourth, half the bulk of the urine, &c. It is also very important that the total quantity of urine passed in the twenty-four hours should be noted.

Bottles used for carrying specimens of urine should be made of white glass, with tolerably wide mouths, and capable of holding at least four ounces; but, if the sediment only of the urine is required, the clear supernatant fluid may be poured off, after the urine has stood in the receptacle for several hours, and the deposit may then be poured into small bottles of an ounce capacity, or even less. The only objection to this mode of collecting urine is, that no idea of the *amount* of sediment belonging to a given quantity of urine can be formed. The bottles may be arranged in a case capable of containing two, four, or six.

309. Importance of Examining the Urine soon after it has been passed, and also at a later period.—In all cases, the urine should, if possible, be examined within a few hours after its secretion, and in many instances, it is important to institute a second examination after it has been allowed to stand for twenty-four hours or longer. Some specimens of urine pass into decomposition within a very short time after they have escaped from the bladder; or the urine may even be drawn from the bladder actually decomposed. It is a mistake to suppose that where bacteria are found in the urine, as it issues from the urethra, or in the fluid drawn off by a catheter, they or their germs have been introduced from without. Bacteria germs exist normally not only in the bladder, but in the kidneys, and will grow and multiply whenever the circumstances become favourable. Sometimes we shall find the secretion highly alkaline, with a strong ammoniacal odour, and containing numerous crystals of triple phosphate, with granules of earthy phosphate; and upon carefully focussing, multitudes of active bacteria will be noticed. In other instances, the urine does not appear to undergo decomposition for a considerable period, and may be found clear, and without any deposit for many days after it has been passed. In those cases in which *uric acid* or *oxalate of lime* is present, we shall find that the deposit almost always increases in quantity after the urine has stood still in the glass for some time. These salts are frequently not discoverable in urine immediately after it has been passed, but make their appearance in the course of a few hours. The deposition of uric acid seems to depend upon a kind of fermentation, which has been studied by Scherer.

In order to obtain sufficient of the deposit from a specimen of urine for microscopical examination, we must place a certain quantity of the fluid in a conical glass (figs. 1, 2, Pl. XV, p. 158), in which it must be permitted to remain for a sufficient time to allow the deposit to subside into the lower part. It is then removed in the pipette (p. 150).

310. Magnifying Powers required in the Examination of the Urine.—Urinary deposits require object glasses varying in magnifying power for their examination, the inch and the quarter of an inch being those most frequently used. The former magnifies about 40 diameters (× 40), the latter from 200 to 220 (× 200, × 220). Large crystals of uric acid may often be readily distinguished by the former, but some crystals of this substance are so minute that it is absolutely necessary to use higher powers. Octahedra of oxalate of lime are frequently so small that they cannot be seen with any power lower than a quarter; and, in order to bring out the form of the crystals, even higher magnifying powers than this are occasionally necessary. Spermatozoa may be seen with a quarter, but they then appear very minute. An eighth of an inch object-glass, which magnifies at least 400 diameters (× 400), will

be of advantage, but I recommend the student to purchase a one-twelfth magnifying about 700 diameters, if he obtains any object-glass higher than a quarter. The casts of the uriniferous tubes, epithelium, and the great majority of urinary deposits can, however, be very satisfactorily demonstrated and studied with a quarter of an inch object-glass. The student should always bear in mind that it is very important to become thoroughly expert in the use of the lower powers before he attempts to work with higher ones.

In some cases, it will be well to subject the deposit to examination in various fluids, such as water, spirit, mucilage, turpentine, Canada balsam, &c., § 74, p. 31.

311. Importance of the Chemical Examination of Urinary Deposits.—In the investigation of those deposits which are prone to assume very various and widely different forms, such as uric acid, it will sometimes be necessary to apply certain chemical tests, before the nature of the substance under examination can be positively determined.

Suppose, for instance, a deposit which is found, upon microscopical examination, not to possess any characteristic form, be suspected to consist of uric acid, or of an alkaline urate, we have only to add a drop of solution of potash, which would dissolve it, and then excess of acetic acid, when the crystals of uric acid will be deposited after some time in their well-known rhomboidal form; or any other chemical tests which should be considered necessary, § 200, p. 167, et seq., may be applied. See also Tables at the end of my work on "Kidney Diseases, Urinary Deposits, and Calculous Disorders."

When it is necessary to resort to chemical reagents, a drop of the test solution is to be added to the deposit which is placed in the cell, or upon the glass slide. If necessary, heat may be applied to the slip of glass by a spirit-lamp, and with a little practice, the student will soon be able to perform a qualitative analysis of a few drops of urine, or of a very small portion of a deposit. See chapter X, p. 154.

312. Examination of the Deposit in the Microscope.—The drop of urine with the deposit, removed by the pipette, being now enclosed in one or other form of cell, §174, pp. 151, 152, various parts of the specimen are to be brought into the field of the microscope. It is better to examine the object as regularly as possible, commencing on one side, and moving it up and down, until the whole has been traversed. After one specimen has been examined, and the nature of its contents noted, another may be treated in the same way. Specimens should be taken from the deposit at different levels; for while some deposits soon sink to the bottom, others are buoyed up, as it were, either by the small quantity of mucus which the urine contains, as is the case with small crystals of oxalate of lime, or by the light flocculent nature of the deposit itself.

As each part of the deposit is brought into the field of the microscope, the student should endeavour to recognize every object as it passes before his eye. This, however, he will find to be for some time a matter of considerable difficulty, arising partly from the great number of deposits which commonly occur together, partly from the very various forms which many of these substances are liable to assume, but chiefly, I believe, from the great number of particles of accidental presence which are found in almost every specimen of urine submitted to examination; more especially in urine obtained from the wards of a hospital, upon which the first microscopical observations are usually made.

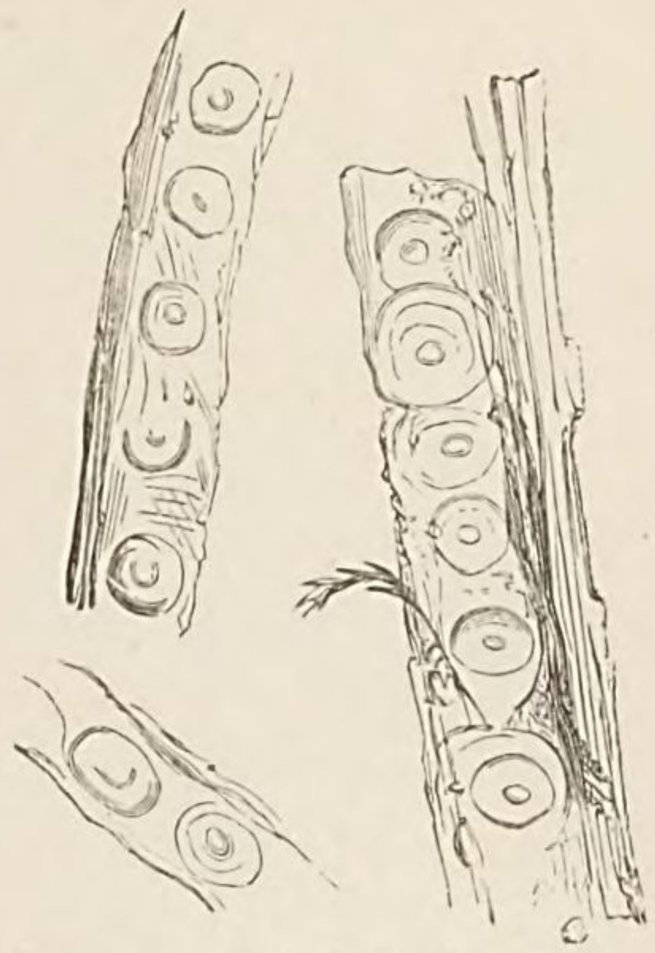
313. Matters of Extraneous Origin frequently met with in Urine.—The substances named in the following list are among those which are very constantly met with amongst urinary deposits, and their general characters are represented in pl. XLVII. Fragments of human hair: cat's hair: hair of different colours from blankets: portions of feathers: fibres of worsted, and fibres of cotton of various colours: fibres of flax: potato starch, rice starch, wheat starch, bread crumbs: fragments of tea leaves, or separated spiral vessels and cellular tissue: fibres of coniferous or other wood swept off the floor: particles of sand: oily matter, in distinct globules, arising from the use of an oiled catheter, or from the accidental presence of milk or butter. Besides the above, there are many other things met with less frequently, as, for instance, fragments of silk, mustard flour, cheese, small portions of the skin of potato, or of different kinds of fruit, and some others which will occur to the mind of every one. With the microscopical characters of these bodies, the student should make himself perfectly familiar as soon as possible; and, as they can be obtained without the slightest difficulty, there is no excuse for ignorance of the general characters of these common things. If he is not able to recognize the ordinary extraneous matters, the student will frequently find himself in considerable difficulty, and his ignorance may lead him to make the most ludicrous and unfortunate mistakes. The origin of most of these substances is so obvious that it need not be stated, but it should be remembered that many of them become slightly altered by standing for some time in the urine.

Fibres of Deal from the Floor.—The only matter of extraneous origin which requires to be particularly noticed, is one which may very easily be mistaken, and, indeed, frequently has been mistaken for tube casts. The substance to which I refer consists of the delicate fibres of coniferous wood which are swept off the deal floor, and thus get into the urine, pl. XLVII, fig. 2. The fibres become soft and swollen by soaking, and sometimes really look very much like large tube casts. The round pores which they contain somewhat resemble epithelial cells. The

Fig. 1.

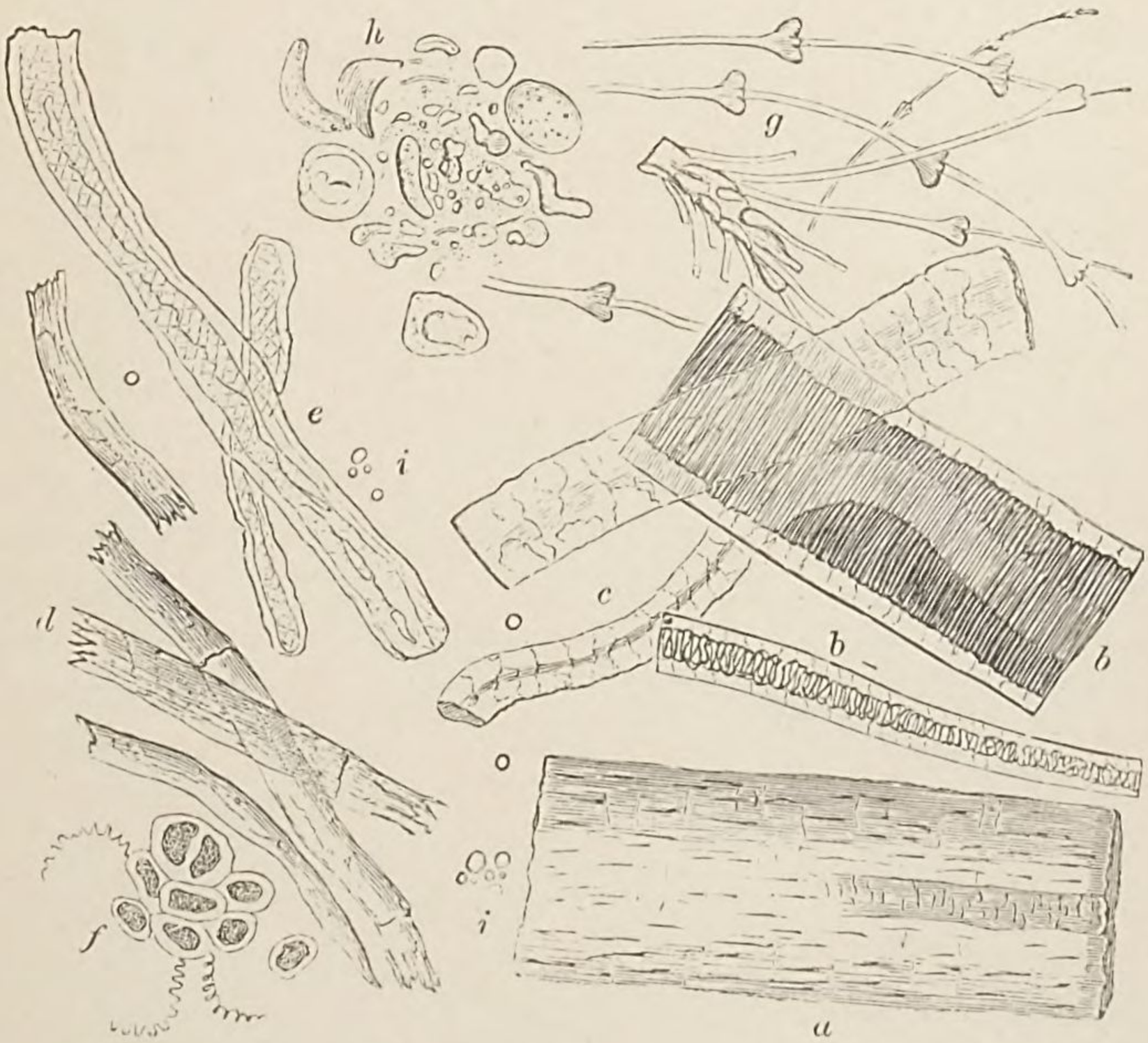
Globules of potato starch, as they appear when examined in water. × 215. p. 314.

Fig. 2.



Small fragments of deal wood swept from the floor. They may very easily be mistaken for casts, and the 'pores,' characteristic of coniferous wood, are not unlike epithelial cells.
p 314

Fig. 3.



a, fragment of human hair. *b*, cat's hair. *c*, hair from blanket. *d*, fibres of flax. *e*, fibres of cotton. These as well as worsted fibres are often coloured. *f* fragments of tea leaves, showing cells and spiral vessels. *g*, portions of feather. *h*, bread crumbs, showing wheat starch partly altered by baking and maceration. *i*, free oil globules, consisting of small particles of butter. All × 215. p. 314.

3000th of an Inch, × 215.

[To face page 314

bodies in question will only be met with when the floor is of deal and often swept. I have found them in very many specimens of urine obtained from King's College Hospital.

It is impossible, as a general rule, to prevent various extraneous matters from falling accidentally into the urine. In wards of hospitals, where the floors are constantly swept, the disadvantage is greatly increased. The practitioner will be surprised at some of the bodies which have been found in urine. Young pediculi are not uncommon, and I have had several specimens brought to me containing the living larvæ of the blowfly, and the pupa of this and other flies. Though it was affirmed that these things were actually passed in the urine, there can be no doubt that they accidentally fell in after the secretion was voided. Much inconvenience arising from the presence of extraneous matters would be prevented if each vessel were provided with a light simple cover. For further remarks on the subject of extraneous matters in the urine, the reader is referred to a paper "On the Characters of Extraneous Matters," in the Microscopical Journal, No. II, and to the plates in my work "On Kidney Diseases, &c."

Substances of various kinds are not unfrequently added to the urine for the purpose of deceiving the practitioner. With this view, hysterical patients sometimes try to impose upon, and excite the commiseration of the physician by adding flour, sand, brickdust, and other powders to the urine. Milk is very commonly added. Such a specimen is very easily distinguished from one of chylous urine by the presence of the numerous oil globules.

In one case which came under the notice of my friend, Dr. Stewart, jeweller's rouge (peroxide of iron) had been added to the urine. The man had been to several of the metropolitan hospitals, and had imposed upon the physicians, but at last Dr. Stewart was able to ascertain the nature of the peculiar red brown deposit. Microscopical Journal, No. II, p. 93.

Dr. Beigel brought me a specimen of urine with a bulky light brown deposit, which was found to consist entirely of yeast which had been added to the urine.

Of Urinary Deposits.

314. Arrangement of Urinary Deposits.—The following arrangement of urinary deposits is based simply upon the appearances which may be observed by the unaided eye. Although such an arrangement must be somewhat artificial, it may help to connect in the student's mind the general appearances of different deposits with their microscopical characters. The proposed arrangement has no reference to the chemical nature, microscopical characters, origin, or to importance in diagnosis of

the several urinary deposits. It is not to be regarded as a scientific classification ; but that it has some practical advantages, will, I think, be admitted.

Upon taking a superficial glance at the more common forms of urinary deposits, it will be noticed that while some are transparent, light, and flocculent, others present the converse of these characters. There are several granular or crystalline substances which form a small dense sediment which sinks to the bottom of the vessel, leaving a perfectly clear supernatant fluid. Deposits will, therefore, be divided into three classes, according to the general characters which they exhibit to the unaided eye.

1. Light and Flocculent Deposits, usually Transparent, and occupying considerable volume.—Mucus, with epithelium of different characters, spermatozoa, vibriones, certain forms of fungi, various forms of casts of the uriniferous tubes, and certain matters of extraneous origin.

2. Dense and Opaque Deposits, occupying considerable bulk.—Urate of soda, pus, phosphates, and certain matters of extraneous origin.

3. Granular or Crystalline Deposits, occupying a small bulk sinking to the bottom, or deposited upon the sides of the vessel.—Uric acid, oxalate of lime, small quantities of triple phosphate, cystine, carbonate of lime, blood corpuscles, &c., with matters of extraneous origin.

To illustrate this chapter, I have selected a few of the drawings of urinary deposits from my work. For further examples I must refer to the plates in "Kidney Diseases, Urinary Deposits, and Calculous Disorders."

First Class of Urinary Deposits.

315. Mucus.—If healthy urine be allowed to stand for a few hours after it has been passed, a bulky, flocculent, and very transparent cloud will be deposited towards the lower part. Upon examining this in the microscope, a few delicately-granular bodies rather larger than a blood corpuscle, will be observed sparingly scattered through a clear and perfectly transparent substance, in which only a few minute granular points and irregular lines can be detected. Mucus is, in fact, the débris from the epithelial surface, and of cells formed within the follicles of the mucous membrane. A few cells of epithelium from the bladder, or from some part of the urinary mucous membrane, are not unfrequently met with. Nothing more is observed in mucus from healthy urine. In some other diseases, however, this mucus increases in quantity, and forms a thick transparent deposit, containing numerous particles similar to those above referred to, with much epithelium, the character of which depends upon the particular part of the mucous membrane affected ; fig. 1, pl. XLVIII, represents the general appearance of mucus found in urine under a magnifying power of 200 diameters. In the upper part of the figure is represented a cell of bladder epithelium.

The mucus which is deposited from many specimens of urine, often

contains a great number of octahedral crystals of oxalate of lime, frequently so very minute as to appear under a power of 200 diameters, like a number of dark square-shaped spots. Their crystalline form may be demonstrated by the use of a higher power, but they may be recognized with certainty with a little practice, as their square shape presents a characteristic appearance which soon becomes familiar to the eye. They are insoluble in a solution of potash, and also in strong acetic acid. These crystals are not commonly deposited until after the urine has left the bladder, and if it be allowed to stand for a long period, they frequently undergo a great increase in size. Upon examination, fragments of hair, small portions of cotton fibre, and other substances of accidental presence, are not unfrequently found to be encrusted with these minute crystals. Oxalate of lime is often deposited in the urine of persons in good health.

A very thick, glairy, gelatinous deposit, which is frequently found in the urine in cases of disease of the bladder, must not be mistaken for mucus. This consists of pus altered by the action of carbonate of ammonia, which has been set free in consequence of the decomposition of the urea by the mucus or some other animal matter acting upon it as a ferment, § 316, after it has left the bladder. In some cases this change commences in the bladder itself, and the expulsion of the viscid glairy mass often occasions great pain, and sometimes cannot be removed without great difficulty. Urine of this kind exhibits a highly alkaline reaction, evolves an ammoniacal odour, and frequently contains a considerable deposit of crystals of the triple or ammoniaco-magnesian phosphate, with granules of phosphate of lime. Liquor ammonia and potash exert a similar change upon pus out of the body.

316. Vibriones, or Bacteria.—After mucus has been allowed to stand for some time in urine, and occasionally in the mucus while it yet remains within the bladder, numerous vibriones make their appearance. These organisms are seen as minute lines under the microscope, which exhibit very active movements: the longer ones twisting about in a serpentine manner. They are frequently developed in urine before it has left the bladder, and always occur in decomposing urine. As in the old epithelial cells of the mucous membrane of the mouth, the germs of bacteria and of certain microscopic fungi are invariably present, we are not surprised to find them in the old cells of vaginal and bladder epithelium, and in the epithelium of other parts of the genito-urinary mucous membrane. Fig. 2 *b*, pl. XLVIII, represents the appearance of some of the commonest vibriones met with in urine. The "Trichomonas Vaginæ," discovered by Donné, is said to be found sometimes in the urine of women suffering from leucorrhœa.

317. Torulæ.—Certain forms of vegetable fungi or torulæ are usually developed in urine after it has been standing some time. The

period which elapses before the appearance of the fungi, and the particular species or the special form assumed by the species which is developed vary much in different specimens of urine, and in different cases of disease. In diabetes, torulæ are often present when the urine is voided, and are developed in quantity very soon (within a few hours) after. The growth of these organisms at this early period leads the observer to suspect the presence of sugar, which must be confirmed by the application of chemical tests. See "Kidney Diseases, Urinary Deposits, and Calculous Disorders."

Fig. 2 *a*, pl. XLVIII, represents sporules of fungi of three different characters. Figs. 3, 6, and 7 show the appearance of fungi often developed in urine. All these were found in acid urine, and uric acid was present in the specimen which contained the fungi represented in the two lower drawings.

318. Sugar Fungus. Penicillium Glaucum.—Dr. Hassall communicated a most interesting paper upon the development of torulæ in the urine, to the Medico-Chirurgical Society, which will be found in the volume of Transactions for 1853. This observer arrived at the conclusion that there is a species of fungus which is developed in specimens of urine, containing even very minute traces of sugar, which may be looked upon as characteristic of the presence of this substance, as it occurs in no other condition of the urine. The sugar fungus, is represented in different stages of growth in fig. 9, after Hassall, and other drawings of it are given in figs. 10, 11. The sugar fungus in diabetic urine is identical with the yeast plant. The sporule state is represented in the upper part of figure 9, and at *a* is shown the thallus of the sugar fungus. The fructification of this fungus is represented in fig. 5.

Besides the sugar fungus, however, there is another species which is very commonly met with in acid urine containing albumen, if exposed to the air. This is the *Penicillium glaucum*. This species is represented in different stages of growth in figs. 3, 4, 6, 7, 8. Its fructification, which is very different from that of the sugar fungus, is shown in fig. 4.

The microscopical characters of the fungi in different specimens of urine vary considerably; but these differences depend not upon the existence of several distinct species of organisms, but are due rather to the stage of development which the fungus has reached, and the condition present during its growth. In one specimen of urine, the growth of the fungus may be arrested at the sporule stage, figs. 2, 9, 10, 11; in another, not until a thallus, figs. 6, 7, 8, 9, is formed, and in a third it goes on until aerial fructification takes place, and new spores are developed. The degree of acidity of the urine, and the length of time during which it has been exposed to the air, appear to determine, in great measure, the stage of development which the fungus attains.

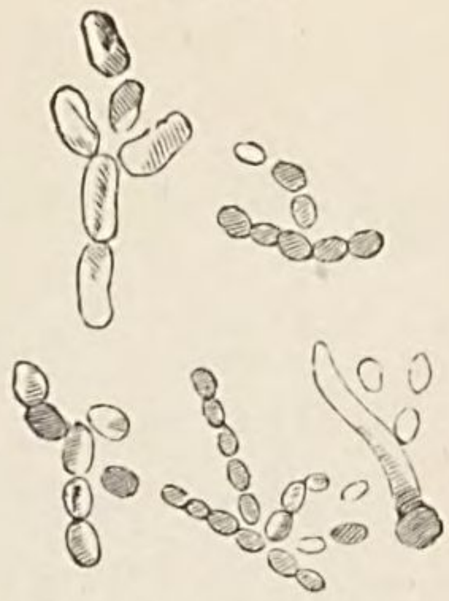
Fig. 1.

Vesical mucus, found in urine. At the upper part of the figure a bladder epithelial cell is represented × 215. p 346.

Fig. 2.

Vegetable organisms met with in urine. *a*, different forms of fungi. *b*, vibriones. × 215. p. 348.

Fig. 3.

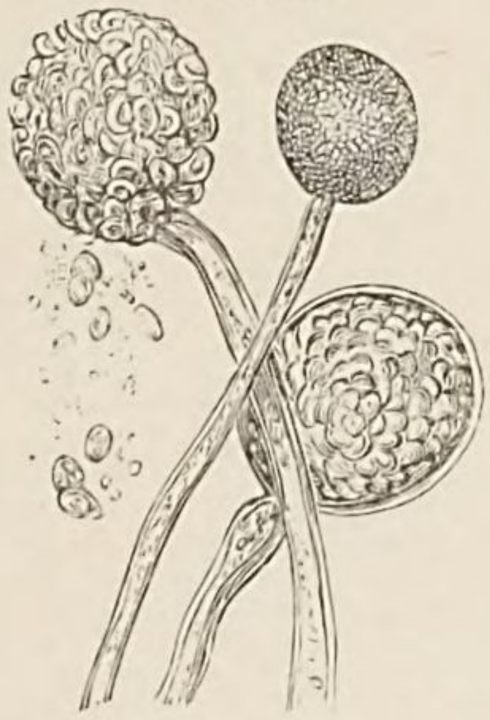


Penicillium glaucum, from acid urine. p. 348.

Fig. 4.

Fructification of penicillium glaucum.

Fig. 5.



Fructification of yeast fungus.

Fig. 6.

Penicillium glaucum. × 215.

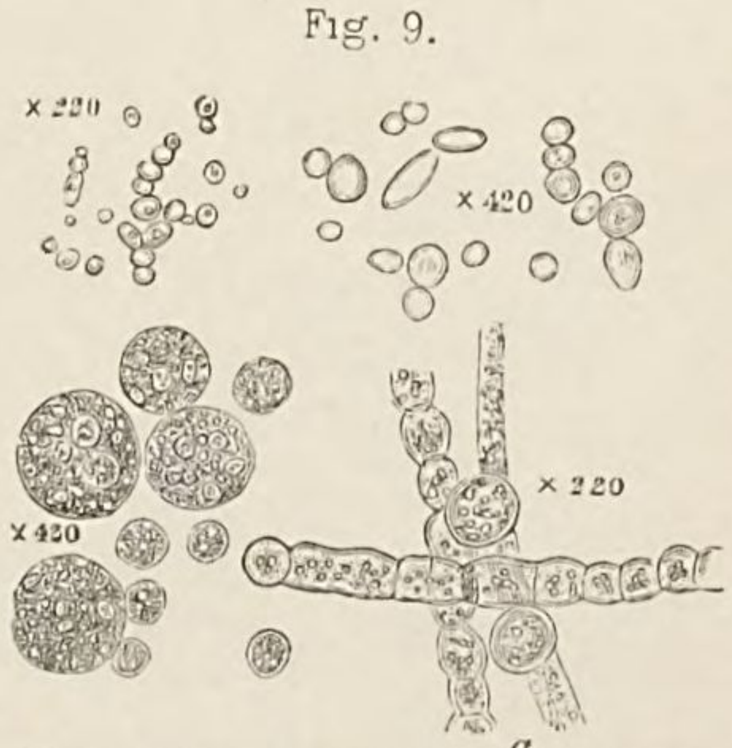
Fig. 7.

Penicillium glaucum, from acid urine. × 215.

Fig. 8.

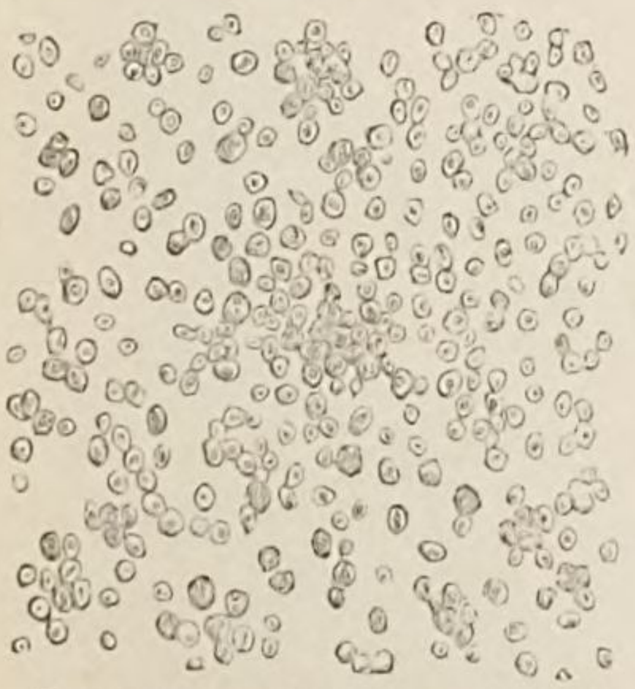
Penicillium glaucum, found in diabetic urine four days after it was passed. × 215. p 348.

Fig. 9.



The sugar fungus, from diabetic urine. After HASSALL. p. 348

Fig. 10.



Yeast torulæ, from beer. × 215.

Fig. 11.

Yeast added to diabetic urine, and allowed to stand in a warm place forty eight hours. Showing growth of the torulæ.
a, × 215. *b*, × 400.

1000th of an inch ,___, × 215
" " |_____| × 400.

[To face page 348

The penicillium glaucum, as well as the sugar fungus, may be met with in saccharine urine, because all the necessary conditions for its development may exist, namely, exposure to air, an acid liquid, a little phosphate, and a certain quantity of nitrogenous matter; but the sugar fungus is found only in those specimens of urine in which to these three conditions is added a fourth, viz., the presence of sugar.

319. Sarcinæ.—These vegetable organisms, not uncommon in the matters rejected in certain cases of obstinate vomiting, are occasionally met with in urine. The specimens occurring in this fluid are usually more minute than those obtained from vomit. The characters of sarcinæ are described in chapter XX. See also page 289.

320. Epithelium of the Genito-urinary Passages.—The forms of epithelium which may occur in urine are very numerous, as the characters of the cells differ very much in different parts of the genito-urinary mucous membrane. The specimens represented in figs. 1 to 6, pl. XLIX, were carefully removed from the mucous membrane of the urinary passages of a healthy male subject, with the exception of a few cells which were found in urine.

Kidney.—Convoluted Portion of the Tubes.—The epithelium is of the variety termed glandular, or secreting epithelium, and forms a single thick layer of cells upon the basement membrane. The characters of this variety of epithelium have been described in page 237.

Straight Portion.—The epithelium is flatter, and approaches more nearly to the scaly variety. It forms a thin layer on the surface of the basement membrane.

Pelvis of the Kidney.—The epithelium consists of flat thin scales, which are united together at the edges without overlapping each other. This is termed tessellated epithelium, fig. 2, pl. XLIX.

Ureter.—The epithelium is very abundant, and of the columnar or cylindrical form. The nucleus is usually large and distinct, fig. 3, pl. XLIX.

Bladder.—The epithelium of the bladder differs much in different parts. In the fundus there is much columnar epithelium mixed with the large oval cells represented in fig. 6, pl. XLIX—whereas, in that termed the trigone, the large flattened cells, with a very distinct nucleus and nucleolus are most abundant. The columnar epithelium appears to line the mucous follicles, while the scaly variety lies on the surface of the mucous membrane between them.

Urethra.—The epithelial cells of the urethra, fig. 4, pl. XLIX, are, for the most part, of the columnar form, but mixed with this, there is also a good deal of scaly epithelium. Towards the orifice, the epithelium is almost entirely of the scaly variety.

Vagina.—The large cells of scaly epithelium, so commonly met with in the urine of females, and derived from the vagina, are repre-

sented in fig. 5, pl. XLIX, and in fig. 8, pl. XXIX, page 232. They vary, however, much in size and form, and are sometimes very irregular in shape, with uneven ragged edges.

321. Spermatozoa.—The urine should be examined for spermatozoa soon after it has been passed, but they are not so rapidly destroyed as has been supposed. They may be detected with a power of about 200 diameters, figs. 8, 9, pl. XLIX, if the eye is familiar with their appearance; but to demonstrate them to persons who have not seen them before, it is better to employ a power of from 300 to 700 diameters.

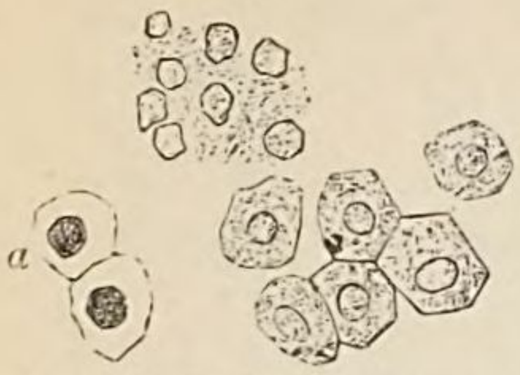
The detection of spermatozoa in the vaginal mucus in cases of suspected rape, is of immense importance. This is one of the cases in which the practical utility of the microscope is quite unquestioned. The mucus may even be dried and remoistened without the forms of the spermatozoa being changed. For cases see "Archives of Medicine," vol. I, pages 48 and 139; also my work on "Kidney Diseases, Urinary Deposits, and Calculous Disorders."

The occasional presence of spermatozoa in the urine is not inconsistent with perfect health. It is only when their appearance is constant and accompanied with important symptoms that the practitioner is justified in interfering. We must always exercise the greatest caution in these cases, for the mere mention of the word "spermatorrhœa" has done more harm to the patient's mind, than can be counterbalanced by the best medical treatment of his body. The occasional presence of spermatozoa in urine must not be looked upon, in itself, as evidence of the existence of that condition, to which the term *spermatorrhæa* has been in too many instances cruelly applied,—a word which I am very sorry to put into print at all; for I doubt if any one word has been productive of more misery than this. Instead of making use of it, we shall be more correct if we discountenance its use altogether, and say that "the patient suffers from such and such symptoms associated with the presence of spermatozoa in the urine." In the majority of cases in which this ill-chosen word is used, it excites terrible alarm in the patient's mind, and perhaps he does not recover for some time,—his notions of the condition having been derived from studying the detestable pamphlets sent round by quacks to frighten silly men and to make dupes of them. In the sense in which it is used by many practitioners, it only means that spermatozoa have been found in the patient's urine, as indeed they occasionally are in the urine of almost every healthy man.

The only bodies at all liable to be mistaken for spermatozoa that I have ever seen, are a form of vegetable growth which I have only once met with, in a specimen of urine kindly sent to me by my friend Dr. Masters. Very careful notes of the case were taken by Mr. C. Roberts, of St. George's Hospital. Some of the bodies in question very closely

PLATE XLIX.

Fig. 1.



Epithelium from the convoluted portion of the uriniferous tube. *a*, treated with acetic acid. p. 349.

Fig. 2.

Epithelium from the pelvis of the human kidney. p. 349.

Fig. 3.

Epithelium from the ureter. × 215.

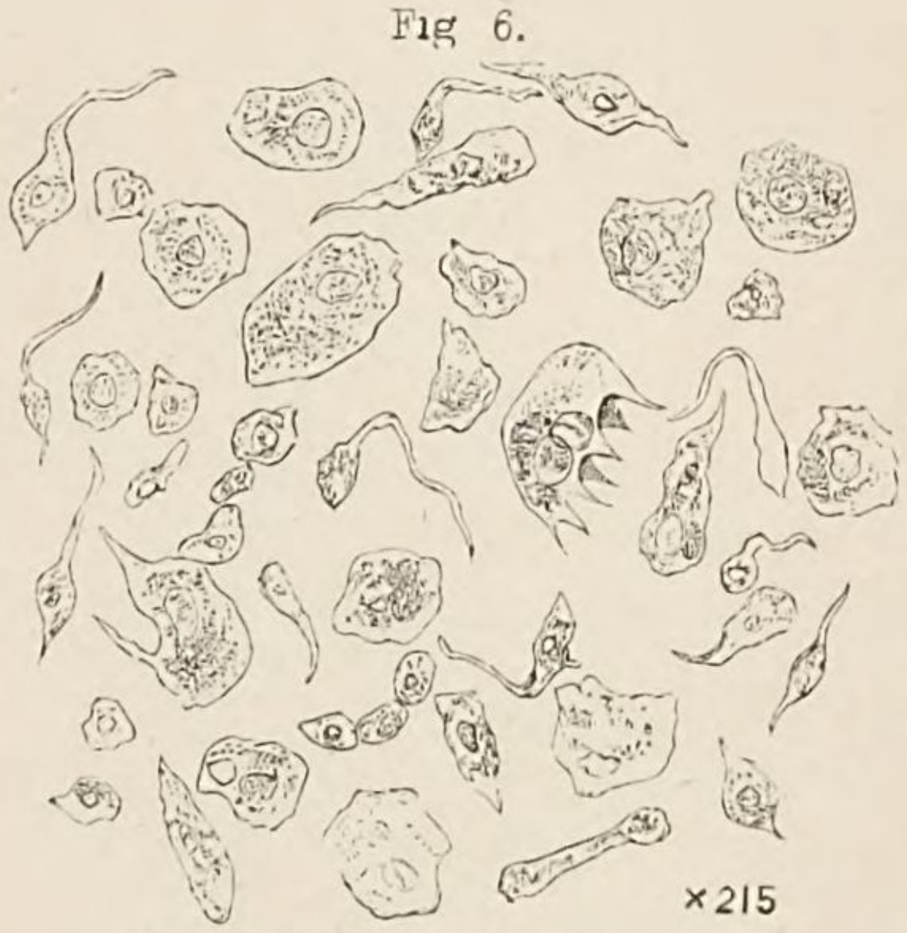
Fig. 4.

Epithelium from the urethra.
× 215. p. 349.

Fig. 5.

Epithelium from the vagina. p. 349.

Fig 6.

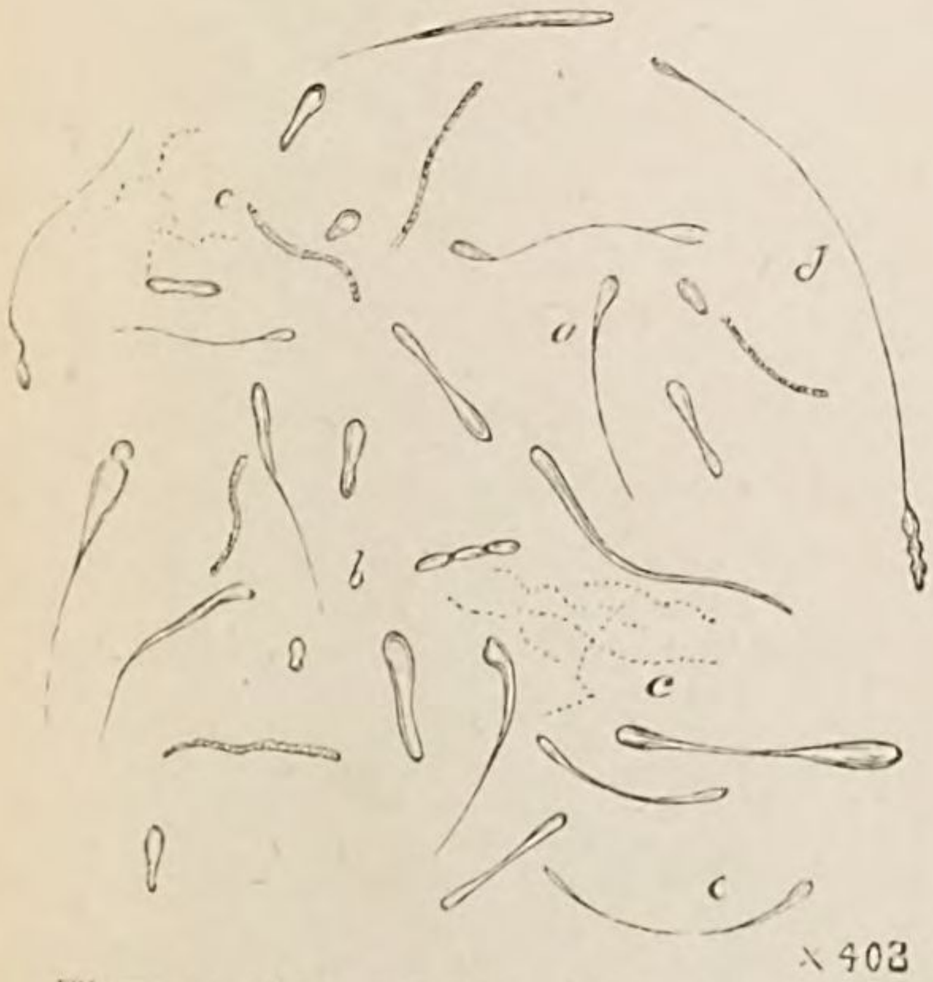


Epithelium from the bladder. p. 349

Fig. 8.

Spermatozoa from urine.
× 215. p 350.

Fig. 7.



× 403

Filaments of a vegetable nature resembling spermatozoa. × 403. p. 350.

Fig. 9.

×215

Cast' of the seminal tubes. Spermatozoa embedded in them. From an old man upwards of 80 years of age.
p. 350.

1000th of an inch ⌊——⌋ × 215.
" " ⌊————⌋ × 400.

[To face page 350

resembled spermatozoa, but their true nature was ascertained by noticing the characters of many other specimens of the vegetable growth. These are figured in pl. XLIX, fig. 7. See "Archives of Medicine," vol. I.

322. Trichomonas Vaginæ.—Donné observed some rounded cells with vibratile filaments projecting from them in the urine of females suffering from leucorrhœa, and considered them to be animalcules. The name *Trichomonas Vaginæ* was applied to them, but subsequent authorities have not been able to confirm M. Donné's observation. Gluge, Valentin, Siebold, and Vogel, consider the so called *trichomonas vaginæ* to be merely a cell of ciliated epithelium from the uterus. Kölliker and Scanzoni have, however, found the trichomonas in the vaginal mucus, both of impregnated and unimpregnated women. (Scanzoni "Beiträge zur Geburtskunde," Band II.) I have never seen cells in any cases at all resembling M. Donné's figure.

Casts of the Uriniferous Tubes.

This to the practitioner of medicine is a most important and interesting class of urinary deposits. The microscopical characters of casts, in different forms of kidney disease, particularly with reference to the diagnosis of pathological changes taking place in the organ, have been investigated chiefly by Dr. George Johnson, to whose observations we are deeply indebted.*

The microscopical characters of the forms of casts most commonly met with will alone be referred to here; and for a full description of the characters of the urine in which they occur, and their pathological significance, I must refer to my work on "Kidney Diseases, Urinary Deposits, and Calculous Disorders."

No conclusion can be based upon the presence of one or two casts of a particular kind, but it is to the general characters of the deposit we must direct our attention. Thus we may find in the deposit from the urine in acute cases which completely, and may be very rapidly, recover, one or two cells containing oil, and one or two casts containing a few oil globules. Now, we must not, from the presence of these, be led into the error of concluding that the case is one of fatty degeneration of the kidney. If, however, there were *numerous* cells and casts containing oil, and these occurred from day to day, such an inference might be justifiable. Nor must we expect to find in any one case *epithelial casts* only, in another *granular casts*, in a third, *fatty casts*, in a fourth, none but *large waxy casts*, and so on; but we must be prepared to meet with several varieties in each case, and must ground our opinion in great measure, upon the relative number of particular kinds of casts,

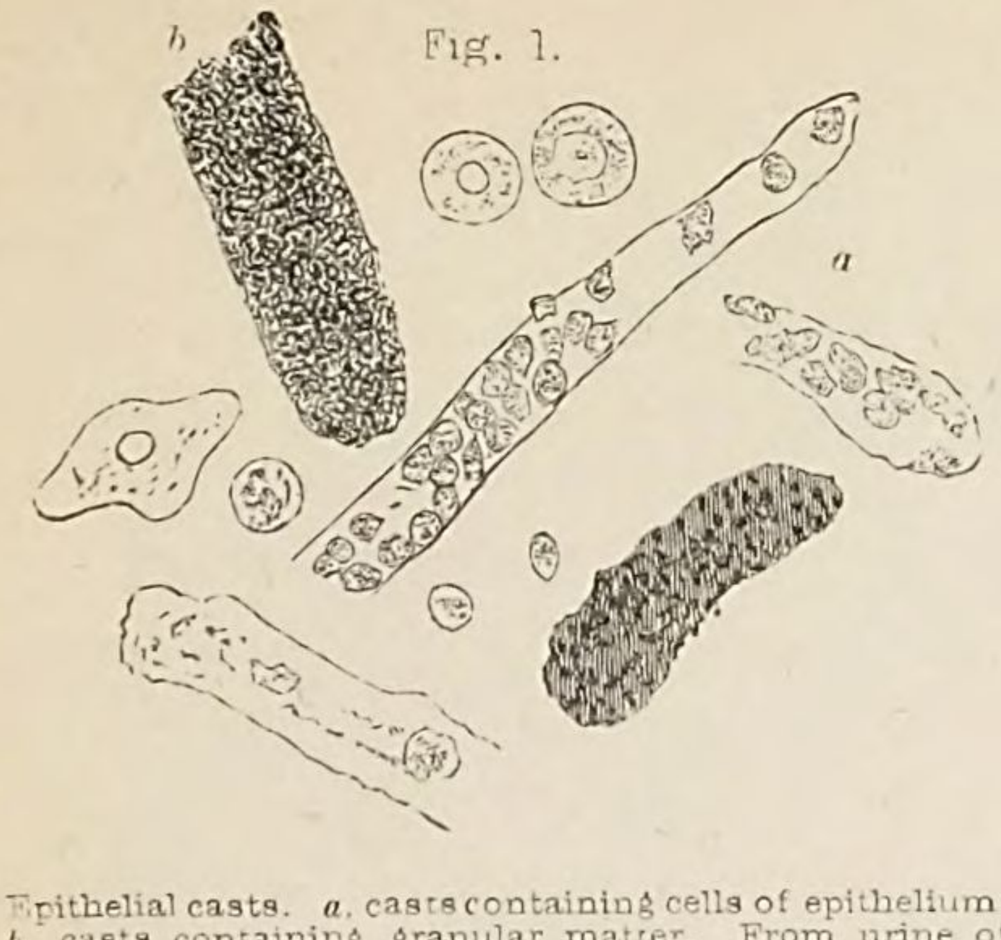
* "On Diseases of the Kidney," 1852.

the constancy of their appearance, and upon the circumstance of other deposits being associated with the casts. For instance, the presence of uric acid crystals and blood corpuscles would render it very probable that the case was acute and of short duration. The absence of these deposits, and the presence of a number of granular or perfectly transparent casts, which can only be seen when the greater part of the light is cut off from the field of the microscope, or the existence of a number of oil casts, render it certain that the disease has been going on for a long time. The first set of facts would indicate that the kidney was in a state of acute inflammation and highly congested; the second that the organs were becoming small and contracted, while the last variety of casts occur when the kidney is of large size, and undergoing fatty degeneration. Such examples might be multiplied.

When we consider how very numerous the secreting tubes of the kidney are, we cannot feel surprised that a different condition should exist in different tubes at the same time. But careful post mortem examinations have taught us that very different morbid appearances are often seen in different parts of the cortical portion of one kidney. It is not difficult, therefore, to account for the fact of the presence of casts differing much in their diameter and characters in the same specimen of urine.

A *cast* is really a mould of a uriniferous tube, and consists of some transparent material which is formed in, or poured out into, the canal, and there rendered firm, entangling in its meshes whatever may be in the tube at the time of its effusion. The cast varies in diameter with that of the central canal; but probably after its formation it contracts slightly, and in consequence it is readily washed out of the tube and escapes with the urine. The diameter of the cast is determined in great measure by the width of the canal of the uriniferous tube, and this varies according to the state of the epithelium. If the epithelial layer lining the tube be of its ordinary thickness, we shall have a cast of medium size. If the cells be enlarged, and adhere firmly to the basement membrane, the cast will be fine and narrow; while on the other hand, if the tube be entirely stripped of epithelium, the basement membrane alone remaining, the diameter of the cast will be considerable. But with reference to the diameter of casts, there is another very important circumstance to be borne in mind:—viz., that as the straight portion of the tubes unite, tubes of large size result, and near the openings at the summit of the papilla, casts three or four times the diameter of ordinary tube casts, or even larger than this, may be found. In describing the different varieties of casts, it will be convenient to divide them into three classes, according to their diameter. 1, *Casts of medium diameter;* 2, *Casts of considerable diameter;* and 3, *Casts of small diameter.*

Fig. 1.

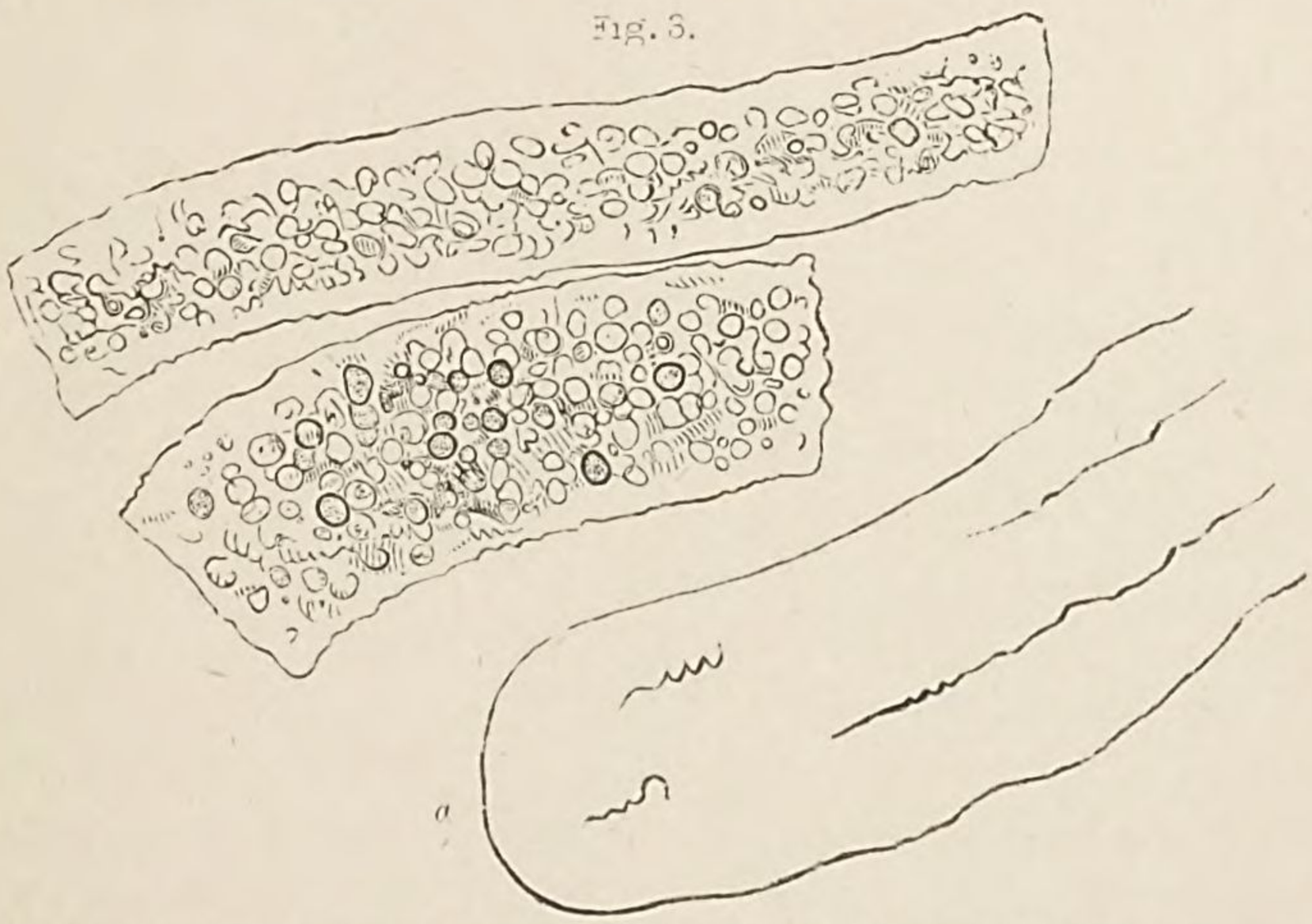


Epithelial casts. *a*, casts containing cells of epithelium *b*, casts containing granular matter. From urine of acute dropsy. p 353.

Fig. 2.

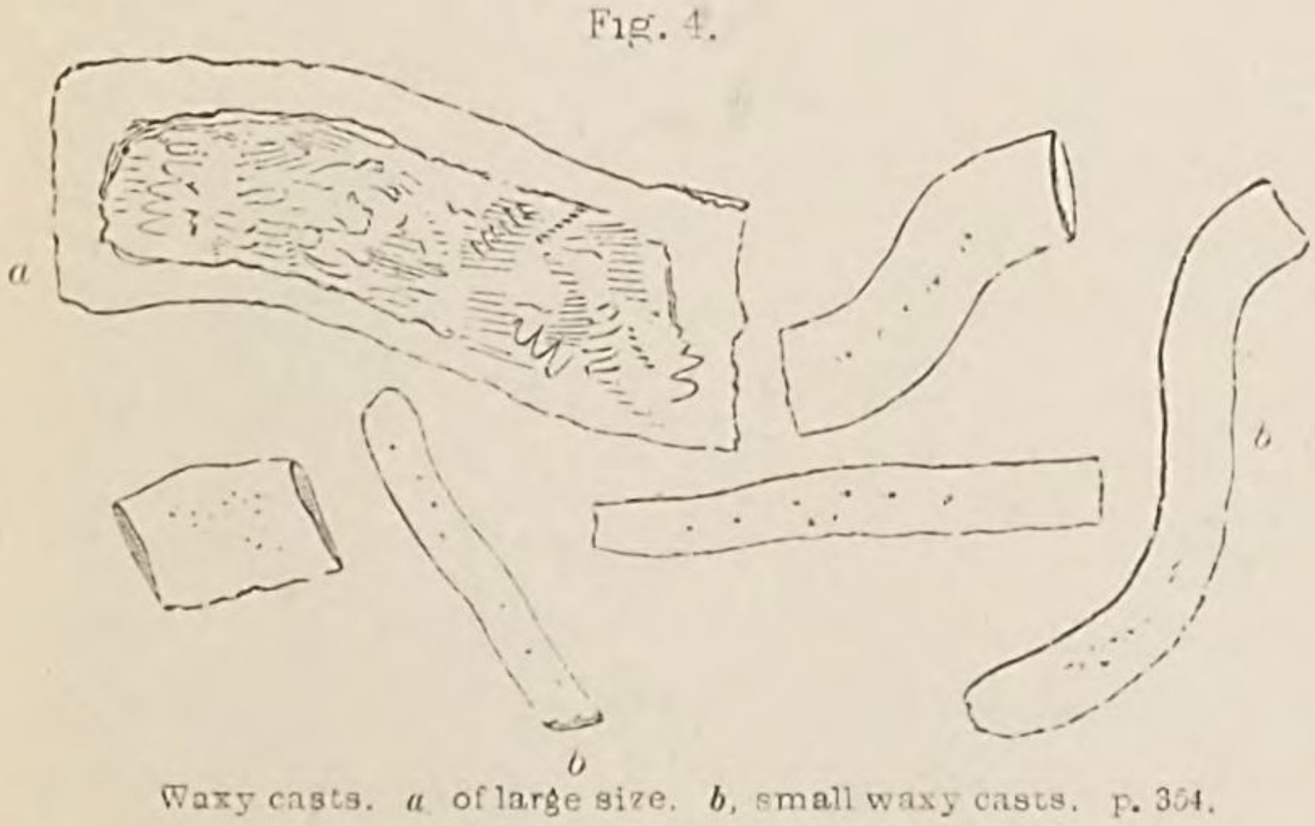
Casts containing fat-cells and oil globules, from a case of fatty degeneration of the kidney. p 354.

Fig. 3.



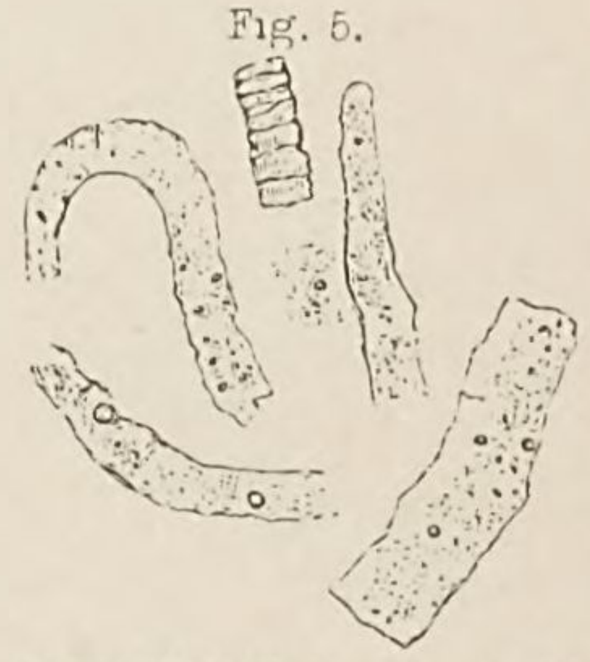
Large casts, some containing many cells; others consisting of a perfectly transparent wax-like material. p. 353.

Fig. 4.



Waxy casts. *a* of large size. *b*, small waxy casts. p. 354.

Fig. 5.



Small granular casts, from the urine of a patient suffering from chronic nephritis. p 354.

1000th of an inch ⌊___⌋ × 215.

All these figures × 215.

References to p. 220.

[To face page 352

CASTS FROM A CASE OF ACUTE INFLAMMATION OF THE KIDNEY.

From the urine of a man, age 45. The patient died comatose three weeks after the first symptoms appeared. There was slight œdema of the legs. The urine contained so much albumen that it became perfectly solid upon being boiled, and after the addition of nitric acid.

a, perfectly transparent wax-like casts. *b*, a very long wax-like cast, consisting of material deposited at two different periods; the original cast in the interior was probably first formed. As this passed down the tube, a new layer of transparent matter was deposited upon the surface. *c*, casts filled with bioplasts looking like pus corpuscles, but larger. *d*, similar bioplasts free in the urine. These existed in considerable number, in fact the greater part of the deposit was composed of these bodies. *e*, a portion of feather. *f*, a piece of cotton fibre. *g*, portion of a human hair. *h*, flax fibre × 215. pp. 353, 354.

$\frac{1}{1000}$ of an inch |___| × 215

[To follow plate L.

Drawings of the various forms of casts, and a description of their characters, and the mode of their formation, will be found in my work on "Kidney Diseases, Urinary Deposits, and Calculous Disorders," but a few of the typical forms have been introduced in pls. L and LI, of this book.

323.—1. Casts of Medium Diameter, *about the* 1-700*th of an inch.* "Epithelial casts" consist of moulds of the tubes in which cells of epithelium are entangled. Some of the cells may be entire, while others are disintegrated, pl. L, fig. 1. Some casts contain only granular matter, figs. 1*b*, 5, and epithelial débris. More rarely casts are met with which contain blood or pus globules. In some instances, entangled in the cast, are numerous oil globules, readily distinguished by their highly refracting nature, with or without cells of epithelium, larger than natural, and gorged with oil, pl. L, fig. 2.

Once I have met with casts of medium diameter, containing well-formed dumb-bell crystals of oxalate of lime. These casts were found in the urine of a patient suffering from cholera. In the same specimen, also, several octahedra of oxalate of lime were present, but these latter were not entangled in the casts, showing that the octahedra were formed after the casts had passed from the kidney. See "Kidney Diseases, Urinary Deposits, and Calculous Disorders."

Occasionally, specimens of urine are met with which contain an abundant flocculent deposit, consisting entirely of casts gorged with cells closely resembling pus corpuscles and free cells of the same character. Such cases are frequently of an acute character, and may terminate fatally in a short time (three or four weeks), but this is not invariably so. I have known several children in whose urine such casts and cells were most abundant, recover completely, and one instance came under my notice in which such cells continued present for upwards of six months. We must hesitate, therefore, before expressing a very unfavourable opinion in these cases, and we ought never to ground our prognosis upon the characters of the urine only.

324. II. Casts of Considerable Diameter, *about the* 1-500*th of an inch.*—"Large waxy casts" are perfectly transparent, and have a glistening aspect, somewhat resembling in appearance the surface of wax as it cools after having been melted. Casts of considerable diameter also occur, of a granular character, and one portion of a cast is often granular while the other is transparent, and containing perhaps a few epithelial cells. Large waxy casts are seen in pl. L, fig. 3; at *a* is represented a large cast, perfectly transparent. Two of the casts in the figure, and the one depicted at *a*, fig. 4, appear to be composed of a material in the interior, differing from that which forms the circumference of the cast—an appearance which I have in several instances observed. In some cases it is probable that these casts of large

diameter are formed in the wide part of the straight portion of the uriniferous tube, § 322. Often it is evident that the material is deposited in successive layers, as in fig. 4, *a*. See also several casts represented in pl. LI. Although in some cases the convoluted portion of the uriniferous tube is wide enough to admit of the formation of a large waxy cast, I have never seen an instance where the tubes leading from the cortical to the medullary portion of the kidney were wide enough to permit such a cast to pass through. I think, therefore, that much of the material must have been deposited as the cast, at first very narrow, passed down the lower wide portion of the uriniferous tube.

325. III. Casts of Small Diameter, *about the* 1-1000*th of an inch.*—"Small waxy casts" are formed in cases in which the epithelium manifests no tendency to desquamate (*non-desquamative nephritis*). The diameter of the cast is, therefore, that of the central canal only, fig. 5, pl. L; and, not unfrequently, we meet with the casts of less than 1000th of an inch in diameter, having a perfectly smooth and glistening surface, and without the slightest trace of granular matter, pl. L, fig. 4. These appear perfectly hyaloid, and, in the microscope, present the same general appearance as a piece of the elastic lamina of the cornea.

326. Fat Cells.—Besides the appearance of the fatty matter in casts, and in cells entangled in casts, it is very commonly met with in small collections in the urine without the presence of casts. Altered epithelial cells of the kidney, enlarged and gorged with oil, fig. 2, pl. L, fig. 1, pl. LII, assume these characters. Sometimes they contain a few oil globules, which are well defined, and are seen to be distinct from each other; while, in other instances, the globules are very minute, and so crowded together, that the cell appears perfectly opaque and dark, resembling the so-called inflammatory globules or exudation corpuscles. Occasionally, cells containing oil globules may be derived from some other part of the mucous surface of the urinary passages. Fig. 2, pl. LII, represents the appearance of some epithelial cells, and collections of oil globules taken from the membranous portion of the urethra. These could scarcely be mistaken for the cells and casts met with in the urine in cases of fatty degeneration of the kidney; but at the same time it is important to bear in mind that cells containing oil globules are occasionally met with in cases where the kidney is not diseased.

327. Conditions in which Fatty Matter may be met with in Urine.—Of late, much attention has been paid to the presence of fatty matter in the urine, and it may be of advantage to refer to the various states in which it may be met with in this secretion:

1. Fatty matter may occur in the urine as distinct and separate globules, resembling those which are produced by intimately mixing oil and water with the aid of mucilage, &c., pl. LII, fig. 1, *b*. When fatty

matter occurs in this state only in urine, it is most probable that it has been mixed with the urine after the secretion left the bladder. It may have dropped into the urine accidentally, or it may have been intentionally added for the purpose of deceiving us, or the secretion may have been drawn off with an oiled catheter.

2. Fatty matter occurs in the urine in the form of globules, inclosed within a cell wall, or in casts, as referred to in p. 354. The composition of the fat in these cases is very interesting. I have shown that it contains much cholesterine dissolved in a more fluid fat, from which it may be readily separated in a crystalline form. From the fatty matter contained in cells obtained from morbid structures in other parts of the body, I have also been able to extract cholesterine; and also from some organs in a state of fatty degeneration. See "Archives of Medicine," Vol. I, page 8, and "Kidney Diseases, Urinary Deposits, and Calculous Disorders."

3. In some of the rare instances which occur from time to time, the so-called "chylous urine," the fatty matter is suspended in a state of exceedingly minute division. In a specimen of chylous urine, for which I have to thank my friend Mr. Cubitt, of Stroud, there existed nearly thirteen grains of fatty matter in a thousand of urine. I could not detect any oil globules. The whole of this large qnantity of fatty matter was in that extremely minute state of division which is termed "molecular," in which condition the fatty matter exists in chyle. See pl. XXXIV, p. 256, fig. 9. Upon microscopical examination, the field was seen to be covered with minute molecules, like small dots, oscillating with a quivering motion about each other. In this specimen there were also a few delicately granular spherical bodies present, exactly like those found in chyle. The appearance of this urine, examined with a quarter, is represented in pl. LII, fig. 3.*

When, therefore, distinct *oil globules* are present in urine, they may have been derived from *oil* or *butter*, which had accidentally fallen into the secretion, or they may be due to the admixture of *milk*. When the oil globules are inclosed within a *cell wall*, or *entangled in casts*, the condition may be looked upon as indicative of "*fatty degeneration*" of the kidney, or of the epithelium situated in some other part of the genito-urinary mucous membrane. And where the fatty matter is in a *molecular state*, the case is one of "*chylous urine*."

Second Class of Urinary Deposits.

The three deposits in this class may appear to the unaided eye very much alike; but they widely differ from one another in their micro-

* The case is fully reported, with analyses of the urine, in vol. I of the "Archives of Medicine."

scopical characters, in their behavionr with chemical reagents, as well as in pathological importance.*

328. Pus.—The microscopic characters of pus have been described in pp. 302 to 306. The form of the globules becomes somewhat altered if they have been soaking in urine for a long time, and ultimately they undergo complete disintegration. Fig. 4, pl. LII, shows the appearance of pus globules; at *a* four are seen which have been acted upon by acetic acid. See also pl. XLIV, p. 304, figs. 2 to 8. If decomposition of the urea, accompanied with the development of carbonate of ammonia, occurs, the globules become converted into a glairy viscid mass: see p. 347.

The mode of formation of pus has been already described (see pp. 247, 249), and it has been shown that pus originates in the bioplasm of cells and tissues in consequence of increased access of nutrient pabulum. The cells of the bladder, urethra, and vagina, very readily assume a condition which results in the development of pus. In fig. 9, pl. LVI, is an excellent illustration of this fact. In this cell the bioplasm was seen dividing and subdividing, and one or two pus corpuscles could be seen in the interior. Such important facts may often be demonstrated in the cells found in the urine in certain cases of disease.

A deposit of pus is very frequently accompanied with crystals of triple phosphate, but this is by no means invariably the case. I have noticed that when the pus is derived from the bladder, the crystals are very frequently present; but in several cases in which large quantities of pus were formed in the kidney, the crystals were altogether absent. This is perhaps to be accounted for by the altered composition of the urine in cases where the kidney is the seat of disease. The phosphate is never derived from the coats of the bladder as was formerly supposed, but is invariably *deposited* from its solution in the urine, either in consequence of alkali being set free from the decomposition of urea, or to double decomposition of certain salts in the urine.

Chemical Characters.—Deposits of pus are rendered clear and glairy by the action of strong alkalies. The mixture is so viscid that it will not *drop* from one vessel into another. The urine in which pus is present contains a trace of albumen, which may be detected by the application of heat, or upon the addition of nitric acid. In certain instances it is a very nice point to decide whether the albumen results from chronic disease, or is due merely to the pus which is present in the urine.

329. Earthy Phosphates.—The earthy phosphates which occur in considerable quantity in urine are *triple phosphate*, or *phosphate of ammonia and magnesia*, and *phosphate of lime*, generally in the form of

* The method of distinguishing these deposits from each other chemically is described in "Kidney Diseases, Urinary Deposits, and Calculous Disorders."

amorphous granules, and small rounded globules, but occasionally in a crystalline form, pl. LV, fig. 5. Triple phosphate (HO,NH_4O,MgO,PO_5), takes the well-known prismatic form. The crystals have obliquely truncated extremities, and a section would be triangular, pl. LII, fig. 5, pl. LV, figs. 1, 4*, 7.

Some of the crystals are more quadrilateral in form, while others appear almost like an octahedron, in consequence of the central part of the crystal not being developed. A crystal of this form is represented in pl. LII, fig. 5. In consequence of the two ends being closely approximated, the appearance of a square crystal, the opposite angles of which are connected by straight lines, is produced. Various modifications of the above forms will also be frequently met with. The faces of the crystals become roughened by standing long in the urine, or, indeed, in pure water, unless a small quantity of some ammoniacal salt be dissolved in it, in which case the crystals will keep unimpaired for a length of time. When triple phosphate is precipitated by the addition of liquor ammoniæ to urine, it occurs as beautiful feathery snow-like crystals, pl. LV, fig. 2. These may be also produced artificially.

The deposit of triple phosphates in urine is always accompanied with phosphate of lime ($2CaO,HO,PO_5$) if the urine be alkaline. This phosphate occurs in the form of small spherical masses or amorphous granules, pl. LV, figs. 3, 4. Often two small globules are joined together so as to resemble a small dumb-bell crystal. The more uncommon modifications of the crystals of earthy phosphate will be considered in the third class of deposits, as they most frequently occur only in small quantity.

Chemical Characters.—Phosphates are soluble in acetic acid, and very readily so in nitric or hydrochloric acid. If ammonia be added to the acid solution, the triple phosphate is precipitated in the form of beautiful stellate crystals, pl. LV, fig. 2, which gradually become altered until prisms are formed, *a*, and phosphate of lime is precipitated in amorphous granules.

330. Urates.—These, the most common of all urinary deposits, are sometimes found in very large quantity. The sediment composed of urates may vary in colour from a pale buff to a tolerably deep red; often, however, it is almost colourless. It is this deposit to which the terms "nut-brown sediment," "lateritious deposit," &c., have been applied, according to the proportion of colouring matter it may contain. It consists principally of urate of soda, with small and variable proportions of urates of ammonia and lime, and traces of urate of magnesia.

Upon microscopical examination it is found to consist entirely of minute granules which are unequally aggregated together in different

parts of the field, pl. LII, fig. 6. More rarely the deposit contains spherical masses of the urate, or small rounded globules, figs. 9, 11. In children, urates are often found in the form of perfectly spherical masses, somewhat resembling in form the crystals of carbonate of lime occurring in horses' urine. Such crystals are figured in "Kidney Diseases, Urinary Deposits, and Calculous Disorders." In the adult also, such spherical crystals are occasionally met with. The late Dr. Kennion, of Harrogate, sent me a specimen of some urine containing the largest spherules of this description that I have ever seen. These are figured in vol. I of the "Archives of Medicine."

The appearance of urate of ammonia artificially prepared, is shown in pl. LII, fig. 9. Fig. 7 shows the appearance of the spherical masses of urate of soda, which form part of the scum of urine while it is evaporating. The smooth semi-transparent flakes consist of phosphates which form a very thin film to which the urates adhere.

Chemical Characters.—Urates are soluble in boiling water, and very soluble in potash. Upon the addition of excess of acetic acid, the soluble urate of potash is decomposed, and after the lapse of a short time, well-formed crystals of uric acid are deposited. These may be examined by the microscope. Urates are entirely combustible at a red heat, and by being treated with nitric acid and ammonia, yield the beautiful purple colour characteristic of murexid (see § 331).

For the method of analyzing these deposits, see Heintz's "Zoochemie," Lehmann's "Physiological Chemistry," vol. i, Bowman's "Medical Chemistry," by Bloxam.

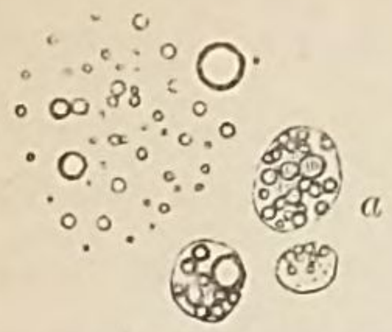
Urate of soda is not unfrequently met with in urinary deposits in the form of small spherical masses, from the surface of which spicules of uric acid project in various directions, pl. LII, figs. 10, 11.

Occasionally, the very dark granular appearance of certain casts is due to the deposition of urates upon their surface and in their substance, after the casts have been passed. Epithelial cells which have been standing for a long time in urine rich in urates, also exhibit the dark granular appearance in every part. Spermatozoa are sometimes invested with a granular covering of urate of soda, and a variety of curious appearances are sometimes thus produced. In these cases the granular appearance is removed upon applying a gentle heat to the slide upon which the deposit is placed, or by the addition of a little dilute potash.

Third Class of Urinary Deposits.

331. Uric or Lithic Acid.—This is one of the most common urinary deposits. Uric acid is frequently deposited after the urine has left the bladder, in consequence of some chemical change, according to Scherer,

Fig. 1.



a, aggregations of oil-globules looking like cells. *b*, free oil-globules as they appear when oily matter has been mixed with urine. × 215. p. 355.

Fig. 2.

Epithelial cells and oil-globules from the membranous portion of the urethra. × 215. p. 354.

Fig. 3.

Fatty matter in a molecular state, as it occurs in chylous urine. × 215 p. 355

Fig. 4.

Pus corpuscles. *a*, treated with acetic acid. p. 356.

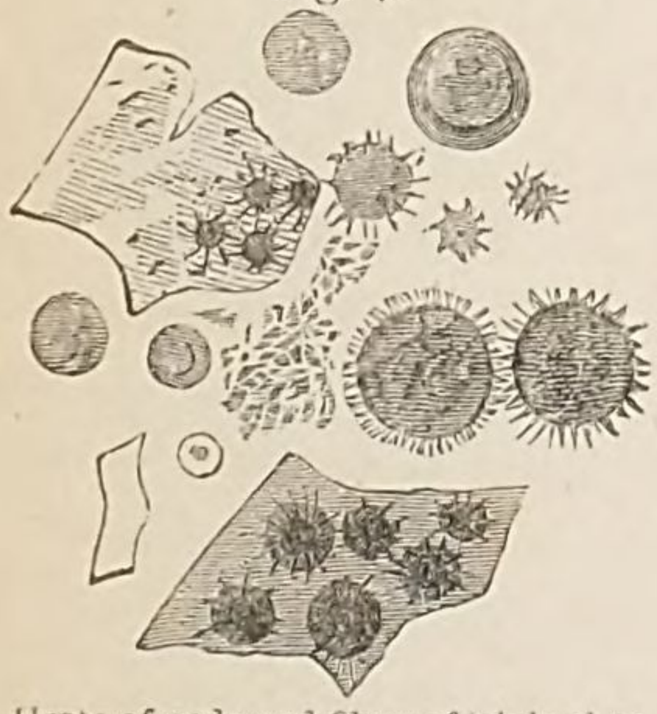
Fig. 5.

Crystals of triple phosphate. Small globules of phosphate of lime with vibriones × 215. p 357.

Fig. 6.

Urate of soda, as it commonly occurs in urine. × 215. p. 358.

Fig. 7.

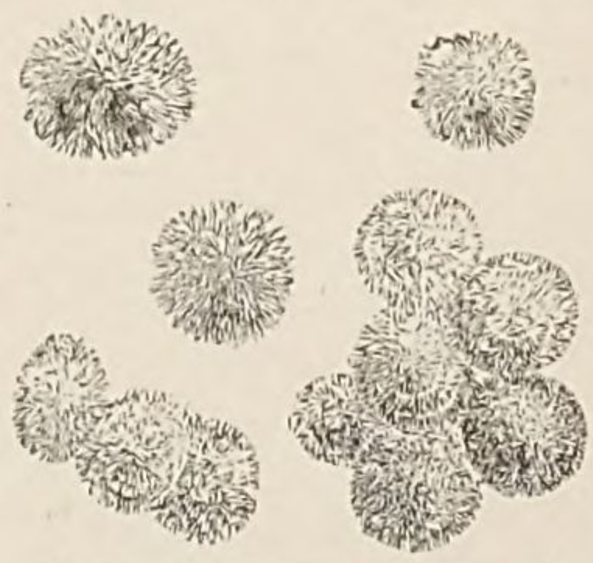


Urate of soda and films of triple phosphate, formed on the surface of concentrated urine. p. 357.

Fig. 8.

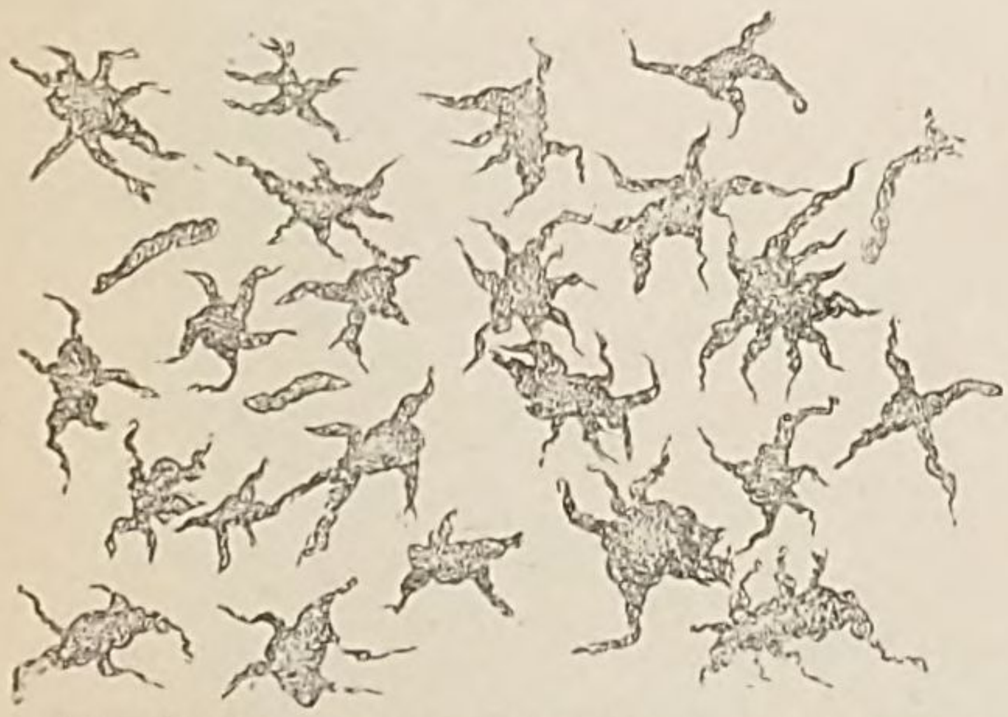
Urate of ammonia, prepared artificially. × 215. p 358.

Fig. 9.



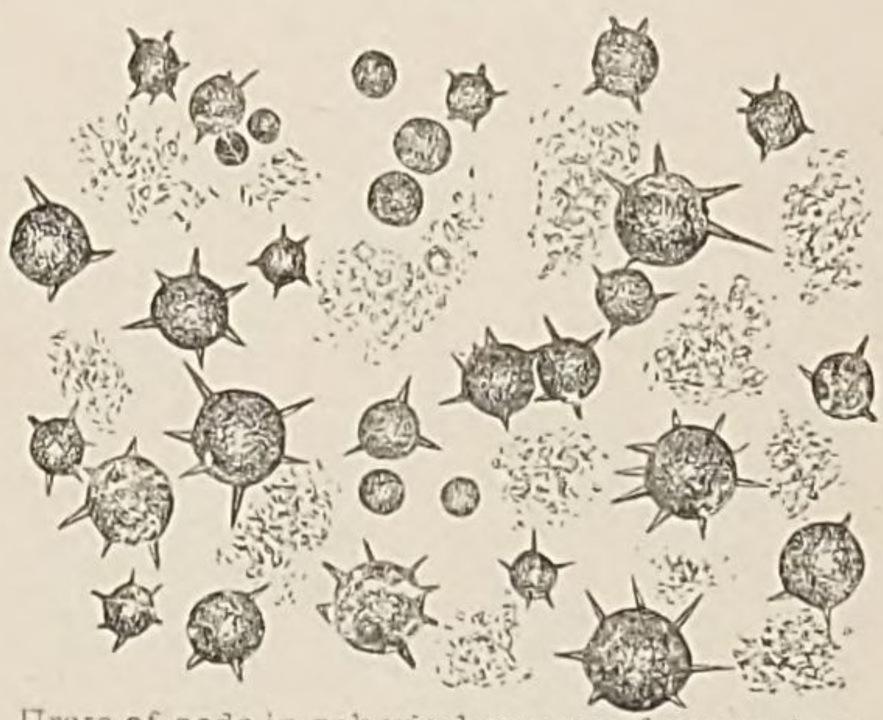
Urate of ammonia, prepared artificially × 215. p. 358.

Fig. 10.



Rare form of urate of soda, from urine of a patient suffering from peritonitis. × 215. p. 358.

Fig. 11.



Urate of soda in spherical masses, from various parts of which minute acicular crystals of uric acid project. × 215. p. 358

1000th of an inch ⌊___⌋ × 215.

[To face page 358

a kind of acid fermentation.* In some instances, however, the uric acid is undoubtedly precipitated before the urine is passed, and occasionally in the secreting structure of the kidney itself. Like the urates, deposits of uric acid vary very much in colour. Sometimes they are nearly colourless, while in other instances, the crystals are arranged in the form of large grains, of a deep red colour—the so-called "Cayenne pepper grains." In pl. LIII, figs. *2*, 6, the appearance of two of these crystalline masses is depicted. The crystals also vary very much in size, so that the deposit may appear to the unaided eye as a granular layer, or as a distinctly crystalline sediment. Deposits of uric acid usually occupy an inconsiderable bulk, compared with that of the urine from which they have been precipitated. Uric acid is occasionally deposited in a granular form. In some cases a very abundant precipitate of minute crystals of uric acid is produced upon the addition of nitric acid to urine. So minute are the crystalline particles formed under these circumstances that the precipitate appears as a flocculent cloud which has been many times mistaken for albumen. Sometimes a firm scum is formed upon the surface of a specimen of urine which, upon examination, is found to be entirely composed of uric acid crystals.

The forms which the crystals assume are very various. The most characteristic, and those most frequently met with, approach the rhomb, and it is in crystals of this character that uric acid is usually deposited when solutions of any of its salts are decomposed by the addition of a stronger acid. Some of the most unlike forms are represented in pl. LIII. Fig. 4 shows the form in which uric acid is often found in the urine of cases of "acute dropsy," and of "dropsy after scarlatina," in which condition it is almost constantly present.

Six-sided crystals of uric acid must not be mistaken for cystine. They may, however, be readily distinguished from these by the fact of two of their sides being longer than the others, and also by their chemical characters. Compare fig. 6*b*, pl. LIII, and fig. 8, pl. LV.

In fig. 1, pl. LIII, are represented some crystals of uric acid, which are occasionally met with. They may often be produced by the rapid crystallization of uric acid in urine, to which nitric or hydrochloric acid has been added. Very many other forms of uric acid are represented in the plates in my work on "Kidney Diseases, Urinary Deposits, and Calculous Disorders."

Chemical Characters of Uric Acid.—A deposit suspected to consist of uric acid, but having no well-defined crystals, may be examined as follows:—A drop of liquor potassæ is to be added to it. If uric acid be present it will be dissolved, and after the addition of excess of acetic acid the alkaline solution will deposit well-formed crystals of uric

* "Untersuchungen," 1843. Lehmann's "Chemistry;" translated by Day, vol. ii, page 408.

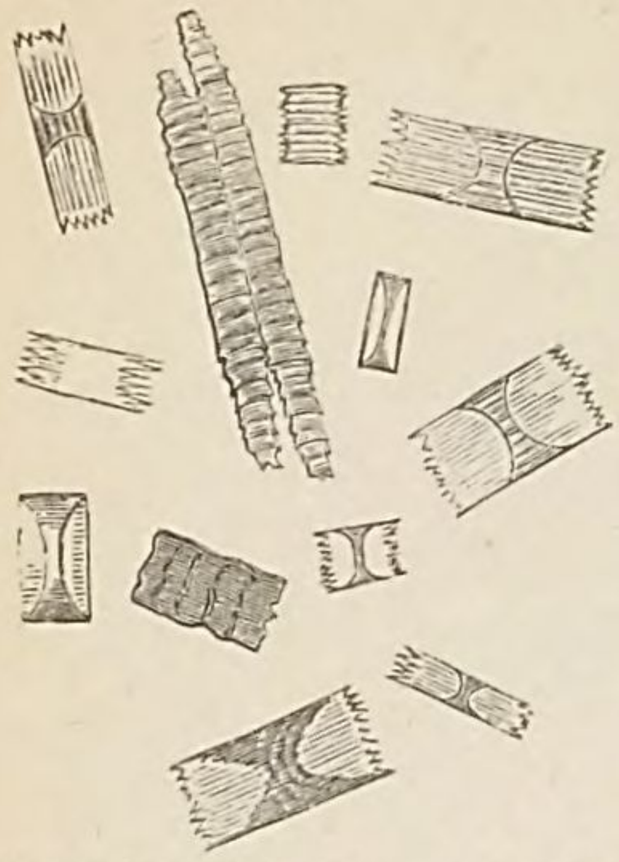
acid. The mixture should be allowed to stand for some time to admit of the formation of crystals. Uric acid is soluble in nitric acid, and if the solution be evaporated to dryness, and a drop of ammonia added, it yields the most beautiful purple colour dependent on the formation of murexide. This is a good test for uric acid when free or in combination.*

332. Oxalate of Lime.—Oxalate of lime was first shown to be a common urinary deposit by the late Dr. Golding Bird. It occurs as a scanty sediment, in which the crystals, if they are large, appear, to the unaided eye, as minute glistening points. Large crystals of oxalate of lime present a beautiful appearance when examined by reflected light, pl. LIV, fig. 9, *d*. If they are subjected to examination in the dry way, they appear like dark cubes, with a clear bright centre, *a*. Their appearance in water and in Canada balsam is shown in the same figure at *b* and *c*. More commonly, however, the crystals do not all sink to the bottom of the liquid, but are, as it were, buoyed up by the small quantity of mucus in which they gradually increase in size. They vary very much in dimensions, from a mere point to a crystal as much as the 1-300th of an inch in diameter.

Oxalate of lime crystallizes in well-defined octahedra, one axis of which is much shorter than the other two. Viewed in various positions, the crystals present a very different appearance, which has given rise to the idea that this substance crystallizes in several different forms in urine. In fig. 7, pl. LIV, several of these appearances are represented; the crystal being the same in each case, but viewed in a different position. In the four lower figures the crystal is shown as it appears when one of

* Uric acid and other substances under certain circumstances give rise to the same reaction with the copper test as sugar, and it has been proved by Dr. De Chaumont that it gives the alcohol reaction with chromic acid, so that in testing urine for alcohol it is always necessary to test not the urine but the distillate. "Almost every urine shows a certain amount of reaction, unless it be first distilled and the distillate operated upon. Even then Dupré found a slight indication of some substance giving the reaction when no alcohol had been taken; but the quantity was too small for examination, and its presence has not been detected by Dr. Parkes in his numerous experiments here. In undistilled urine, the reaction is due (in the absence of alcohol) to uric acid, or any other oxidisable matter that may exist there. Finding in some experiments on myself, that the reaction was present in undistilled urine even sixty hours after any alcohol had been swallowed, I tried the test with what oxidisable substances there happened to be at hand, and found the reaction given with the following:—Uric acid, oxalic acid, oxalate of ammonium, sulphuric ether, benzoic acid, starch, gum-arabic, cane-sugar, and milk-sugar. There was no reaction with pure urea—a non-oxidisable substance. The reaction, therefore, as a test for alcohol, must be looked for in the distillate of urine only, in which case it may probably be depended upon. At the same time, it is possible that any volatile substance swallowed, and capable of being passed off by the urine, might interfere with it; but, as such substances are generally odorous, the smell of the distillate would probably enable the observer to detect them."—(F. De Chaumont, M.D., Army Medical School, Netley.)

Fig. 1.

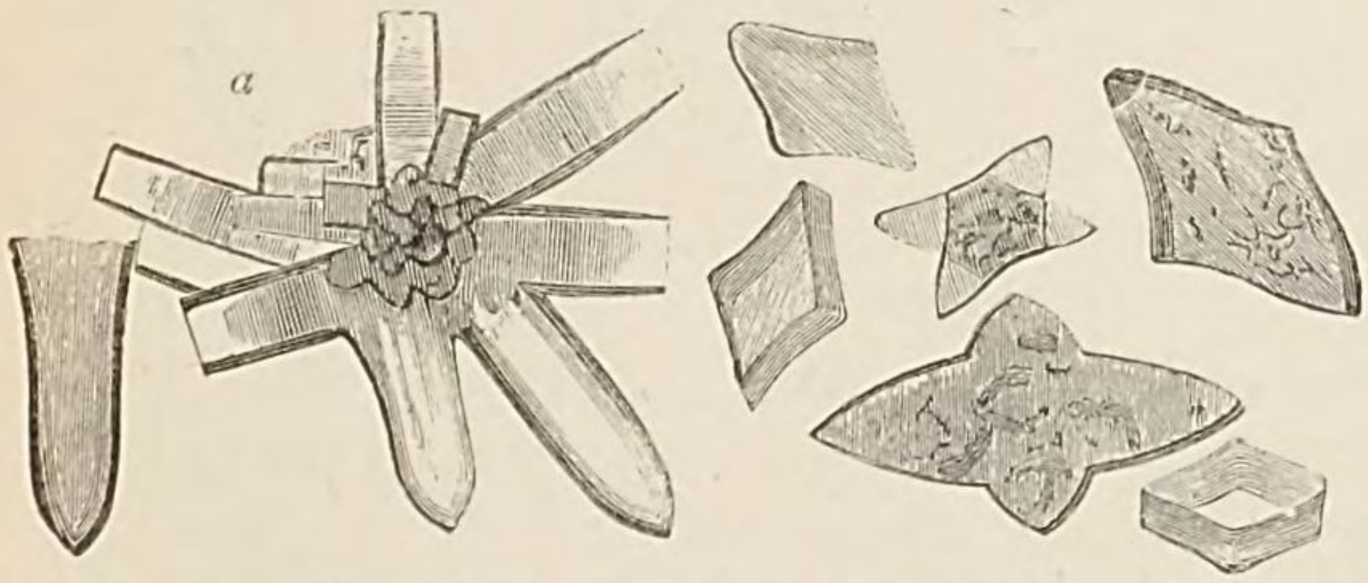


Uric acid crystals, formed by adding nitric acid to urine.
× 215. p 359

Fig. 3.

Uric acid crystals. Human urine. × 130. p. 359.

Fig. 2.

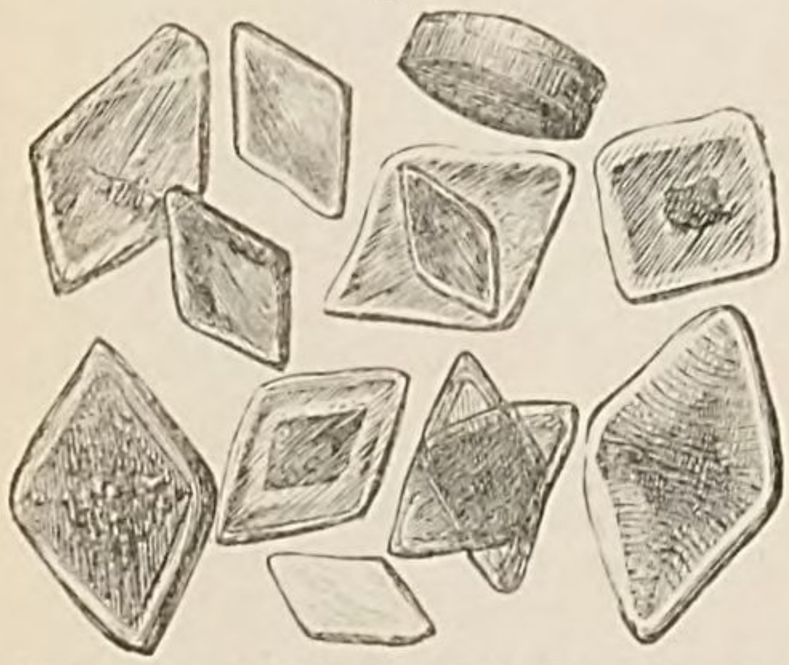


Large halbert-shaped crystals of uric acid *a*, 'Cayenne pepper' grains.
× 215 p 359.

Fig. 4.

Small crystals of uric acid of a rhomboidal form; many of them resemble sections of small cylinders. p. 359.

Fig. 5.

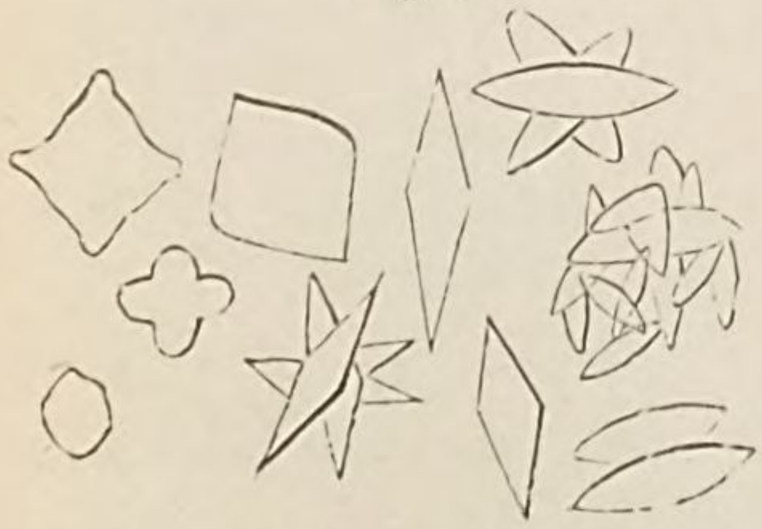


Large crystals of uric acid. Deposited in urine after standing. × 130. p. 225.

Fig. 6.

Less common forms of uric acid crystals. *a*, crystal-like cayenne pepper grains. *b*, six-sided crystals. *c*, mass with small uric acid crystals projecting from it. *d*, small pyramidal crystals of uric acid; very uncommon. *e*, peculiar forms of uric acid. p. 359.

Fig. 7.



Common forms of uric acid crystals p 359.

1000th of an inch ,_, × 130.
" " ,___, × 215

[To face page 360

its lateral angles is towards the observer, and it is rotated upon its long axis. I have been able to observe all these different forms by causing the crystals to turn over in the field of the microscope. I made a little glass model, with the aid of which it was easy to demonstrate the different appearances to the class.

Octahedra of oxalate of lime are frequently deposited after the urine has left the bladder, and may continue to increase in size for some days after their first appearance; so that the urine should be examined, not only soon after it has been passed, but also at a later period.

Not unfrequently the crystals are very minute, and without care in the examination they may be passed over altogether. Minute crystals of oxalate of lime often occur amongst deposits of pale urates, which may obscure them from view. They frequently accompany the crystalline deposit of phosphate of lime, pl. LV, fig. 5. Oxalate of lime is insoluble in potash, so that, when present with urate, the latter may be readily dissolved by the alkali, and the oxalate left perfectly distinct.

Chemical Characters.—Oxalate of lime deposits are seldom met with in sufficient quantity for quantitative analysis. The crystals are insoluble in water, potash, and acetic acid; but soluble in the mineral acids. This deposit, if exposed to a red heat on platinum-foil, becomes converted into carbonate of lime, which effervesces upon the addition of a drop of acid, p. 169.

333. Dumb-bell Crystals.—*Dumb-bell Crystals of Oxalate of Lime.*—These crystals were also first described by Dr. Golding Bird, and proved by him to consist of oxalate of lime; but in consequence of their power of polarizing light, he considered it probable that they might turn out to be composed of oxalurate of lime. This opinion has, however, since been proved to be incorrect. The composition of these crystals is discussed in page 368.

A very perfect form of the dumb-bell crystals is represented in pl. LIV, fig. 8; they were obtained from the urine of a child, two years of age, suffering from jaundice. Besides the dumb-bell crystals, other allied forms are very often present, such as oval and perfectly circular crystals, fig. 5: and not unfrequently crystals of an irregular form occur, one side being even and regular, while the opposite presents different characters. Dumb-bells are usually met with in urine only for a few consecutive days, and they are almost always accompanied with octahedral crystals, fig. 5. I have observed on several occasions that the appearance of the more perfectly formed dumb-bell crystals is preceded and succeeded by the presence of the circular, oval, and less regular forms of crystals, fig. 6.

These crystals are certainly formed in the kidney, for I have seen them in the tubes after death on several occasions, and once I met with them in the transparent casts of the uriniferous tubes which had

escaped in the urine. The crystals take the spherical or dumb-bell form in consequence of the presence of mucus. Carbonate of lime found in the urine of the horse, figs. 1, 2, pl. LIV, and other herbivorous animals, is deposited in allied forms, and the earthy matter of shell, as has been shown by Mr. Rainey, takes a very similar form in consequence of the earthy material crystallising in an organic substance of a colloid nature.

By the prolonged action of acetic acid, I have found that the crystalline matter of the dumb-bell was dissolved, a small quantity of organic matter taking the precise form of the original crystal, and appearing like a cell wall being left, fig. 10, pl. LIV. A similar change takes place in the case of the spherical and dumb-bell shaped crystals of carbonate of lime, so common in the urine of the horse and other herbivora.

The dumb-bell crystals appear to be formed by the aggregation of minute acicular crystals; an arrangement which is well seen in the crystallization of other substances, which, under certain circumstances, assume this form. In fig. 4, pl. LIV, some crystals of urate of potash (prepared artificially) are represented, which crystallized in this form, although the crystalline material was not associated with any form of animal matter. Phosphate of lime also appears to assume the dumb-bell form occasionally. The crystals delineated in pl. LIV, fig. 3, were obtained from the decomposing mucus of the gall-bladder of an ox.

Uric acid occasionally assumes the dumb-bell form; but these crystals are readily distinguished from those of the oxalate of lime by their solubility in solution of potash, and by the difference of their refracting power. See also the observations upon oxalate of lime calculi, page 367.

Chemical Characters.—Dumb-bell crystals possess the same chemical characters as the octahedra of oxalate of lime. They are, however, dissolved by the very prolonged action of acetic acid, fig. 10, pl. LIV. Sometimes aggregations of dumb-bells constituting minute calculi are found in sufficient quantity for analysis.

334. Earthy Phosphate.—Besides the ordinary forms of crystals of phosphate, fig. 5, pl. LII, figs. 1, 2, pl. LV, there are others which occur more rarely in small quantities. Figs. 4, 5, 6, pl. LV, represent some crystals of phosphate, which were formerly considered to be a peculiar form of magnesia, but these crystals have been proved to be composed of phosphate of lime by Dr. Hassall.* This crystalline form of phosphate of lime is often associated with crystals of oxalate of lime, fig. 5. Beautiful crystals of phosphate of lime may be prepared

* "Medical Times," 1851, page 374.

Fig. 1.

Crystals of carbonate of lime, in Canada balsam; seen by transmitted light. pp 362, 363.

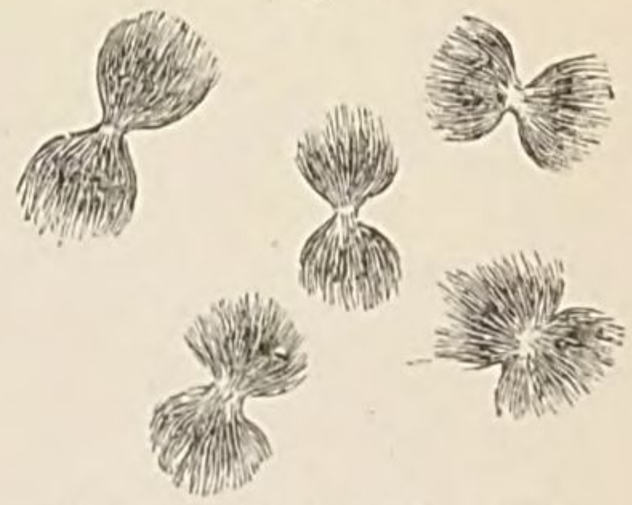
Fig. 2.

Crystals of carbonate of lime, seen by reflected light p. 362.

Fig. 3.

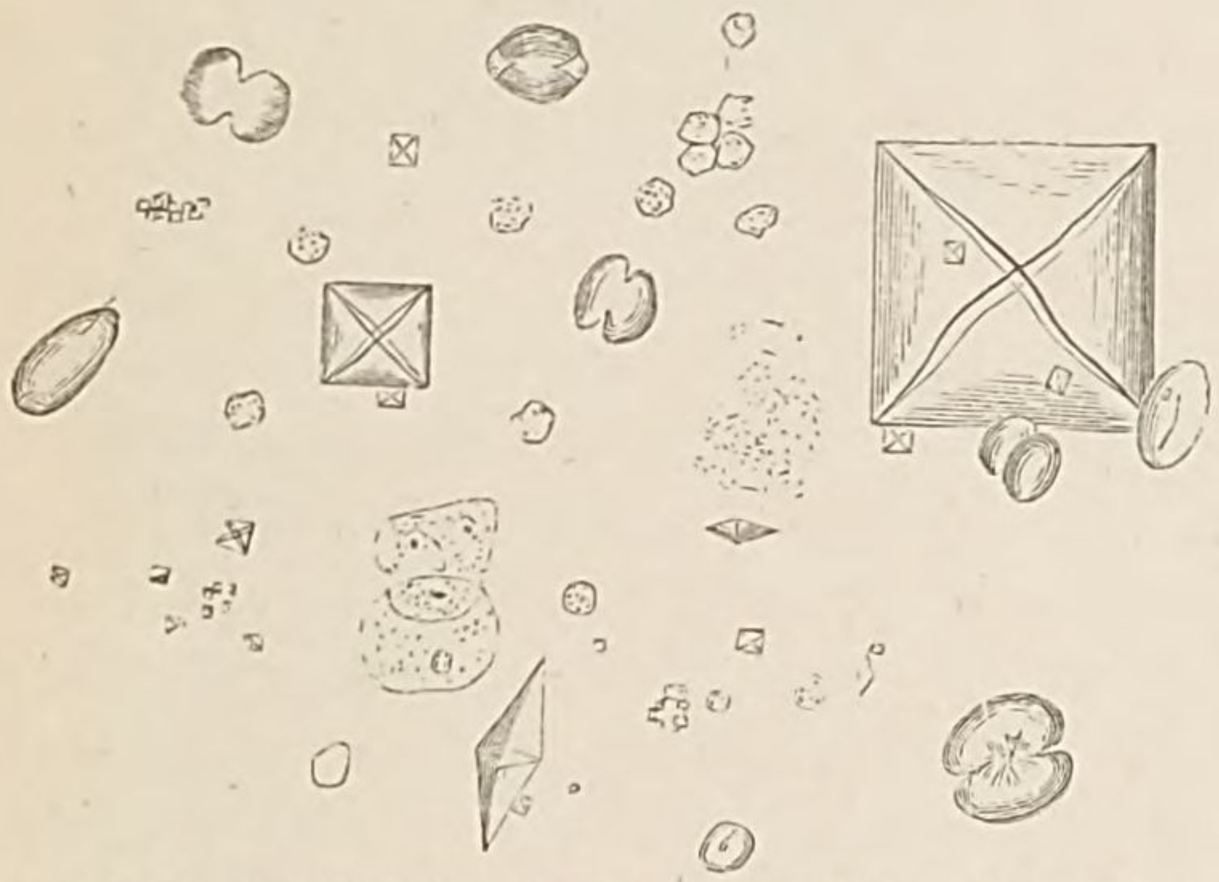
Crystals of phosphate of lime, in the form of dumb-bells. p. 362.

Fig. 4.



Crystals of urate of potash, assuming a dumb-bell form, but evidently composed of acicular crystals. p 362.

Fig. 5.



Dumb-bell and octahedral crystals of oxalate of lime. One very large one is seen at the right-hand side of the figure. pp. 360, 361.

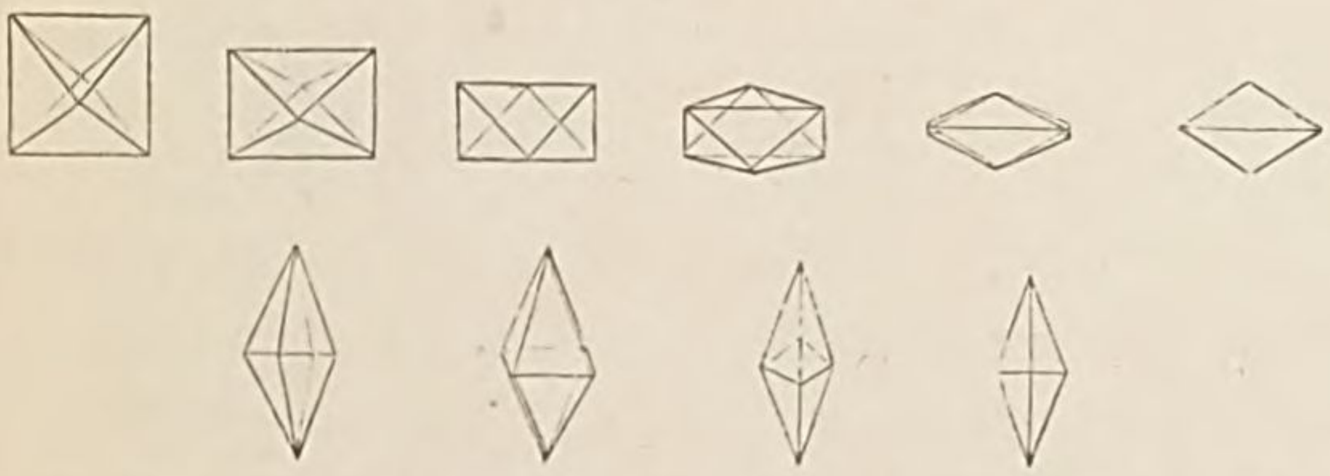
Fig. 6.

Collections of dumb-bell crystals firmly adherent to each other. Such a mass might very easily become converted into a small calculus by deposition of material of a similar composition in the intervals. p. 361.

Fig. 8.

Perfect dumb bell crystal, from the urine of a child two years old, suffering from jaundice. p 361.

Fig. 7.

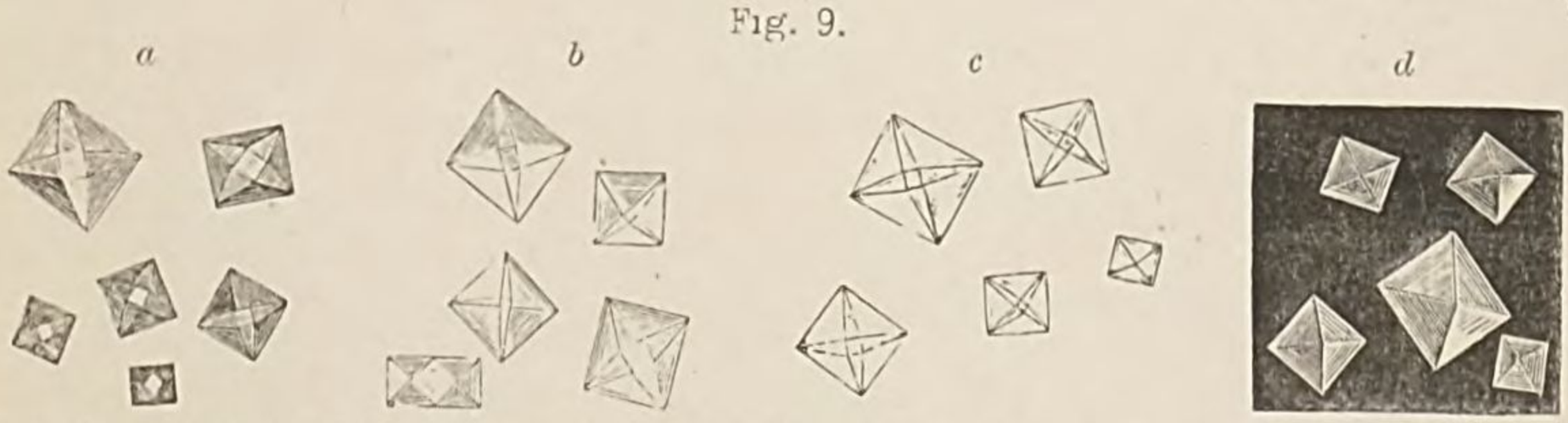


The same octahedron of oxalate of lime, seen in different positions. p. 360.

Fig. 10.

Dumb-bells subjected to the prolonged action of acetic acid, showing the crystalline material nearly dissolved away. p. 362.

Fig. 9.



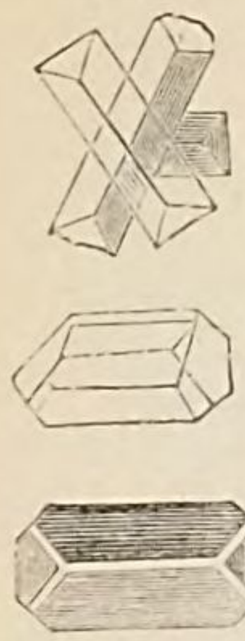
Octahedra of oxalate of lime. *a* by transmitted light in the dry way *b*, in water. *c*, in Canada balsam. *d*, by reflected light. p. 360.

1000th of an inch [___] × 215.

All these figures × 215.]

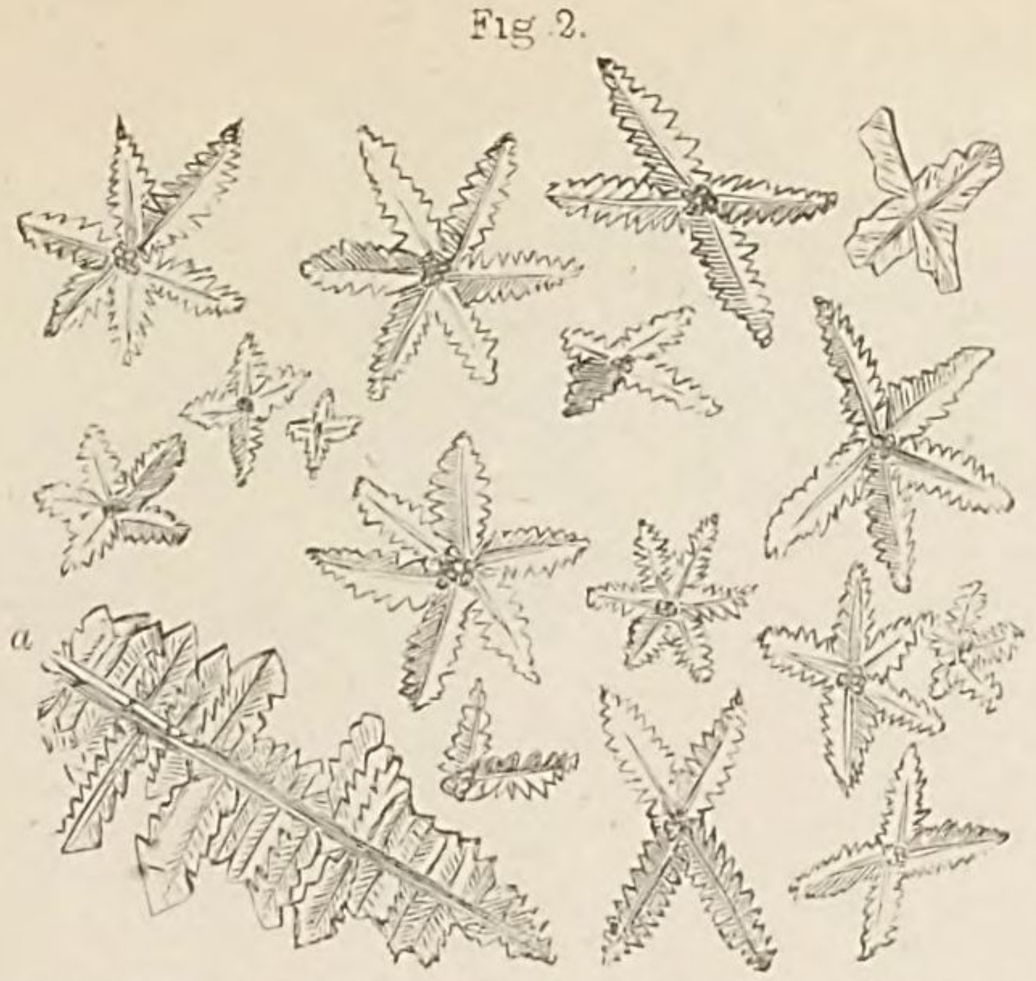
[To face page 362

Fig. 1.



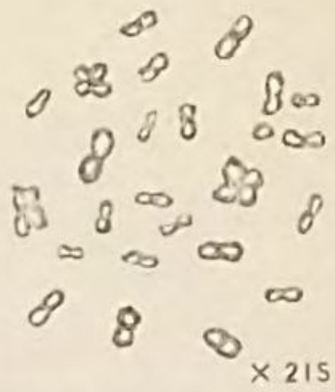
Prismatic crystals of triple phosphate, showing their form. p. 362.

Fig 2.



Stellate crystals of triple phosphate. Formed by the addition of ammonia to urine. × 40 *a* is an arm of a crystal. × 215. p. 357.

Fig. 3.



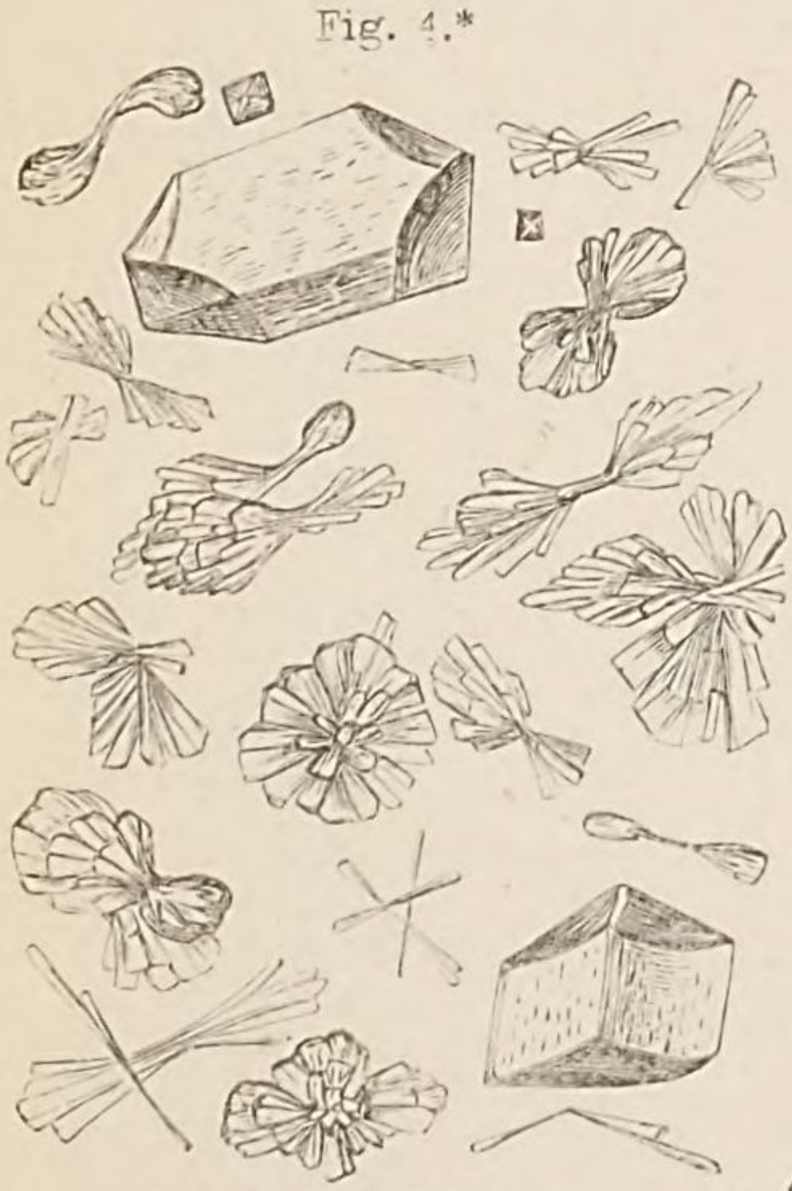
× 215

I ırge granules of phos phate of lime. p 357.

Fig. 4.

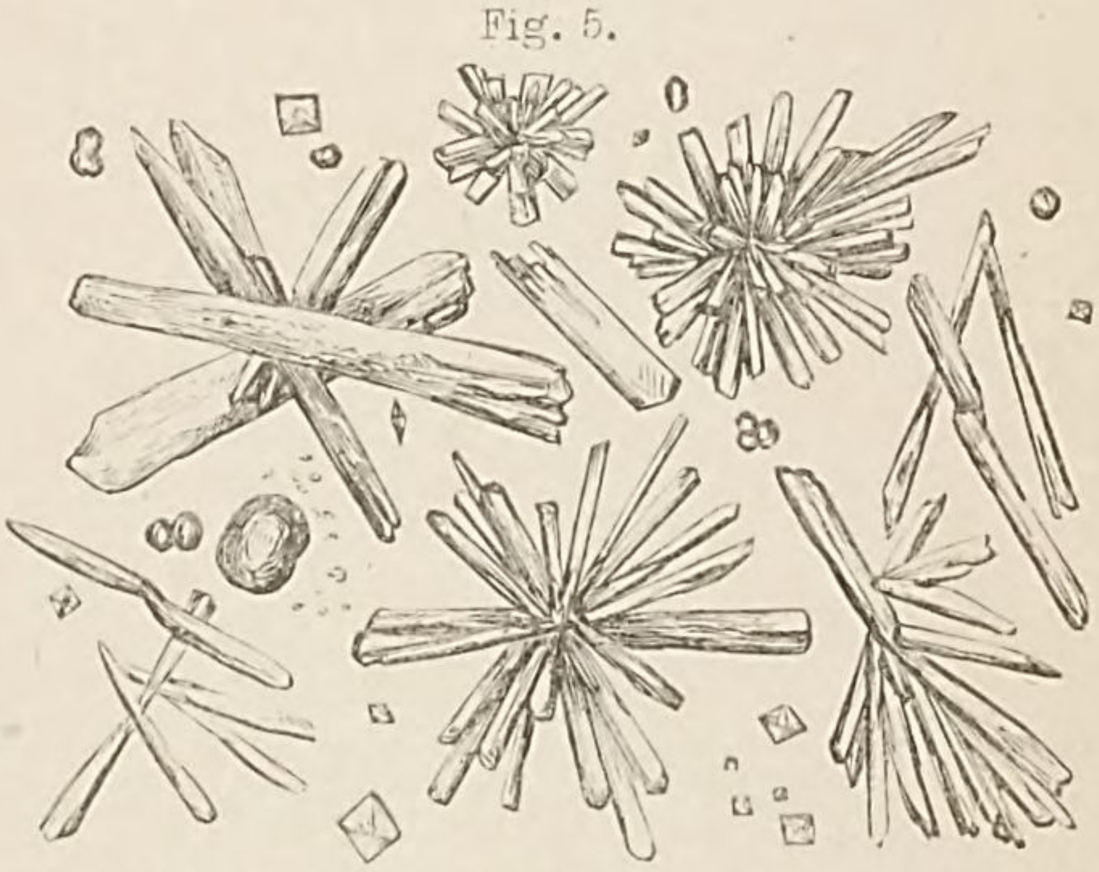
Unusual form of triple phosphate. From the urine of a patient suffering from indigestion in the very hot weather. × 215. p. 357.

Fig. 4.*



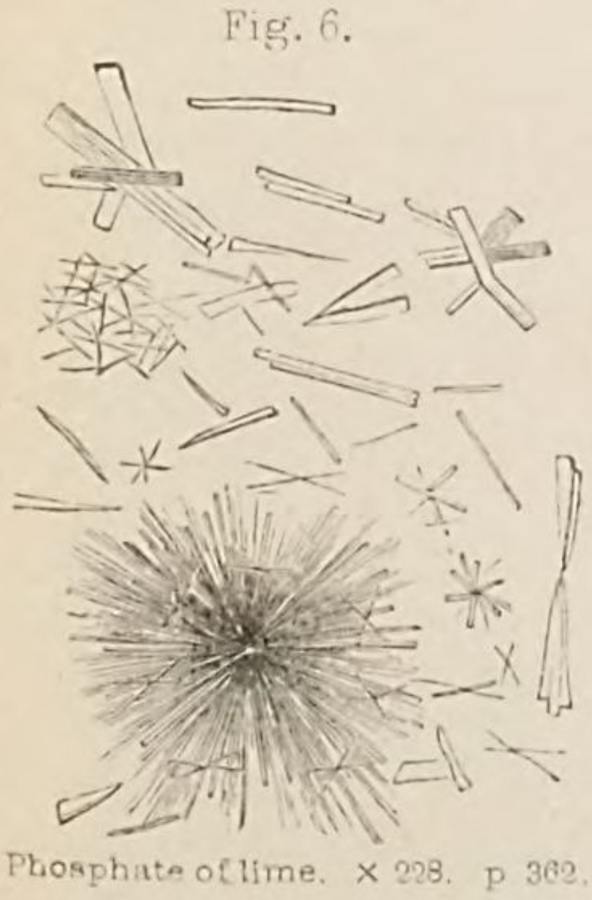
Crystals of triple phosphate. × 130. p 362.

Fig. 5.



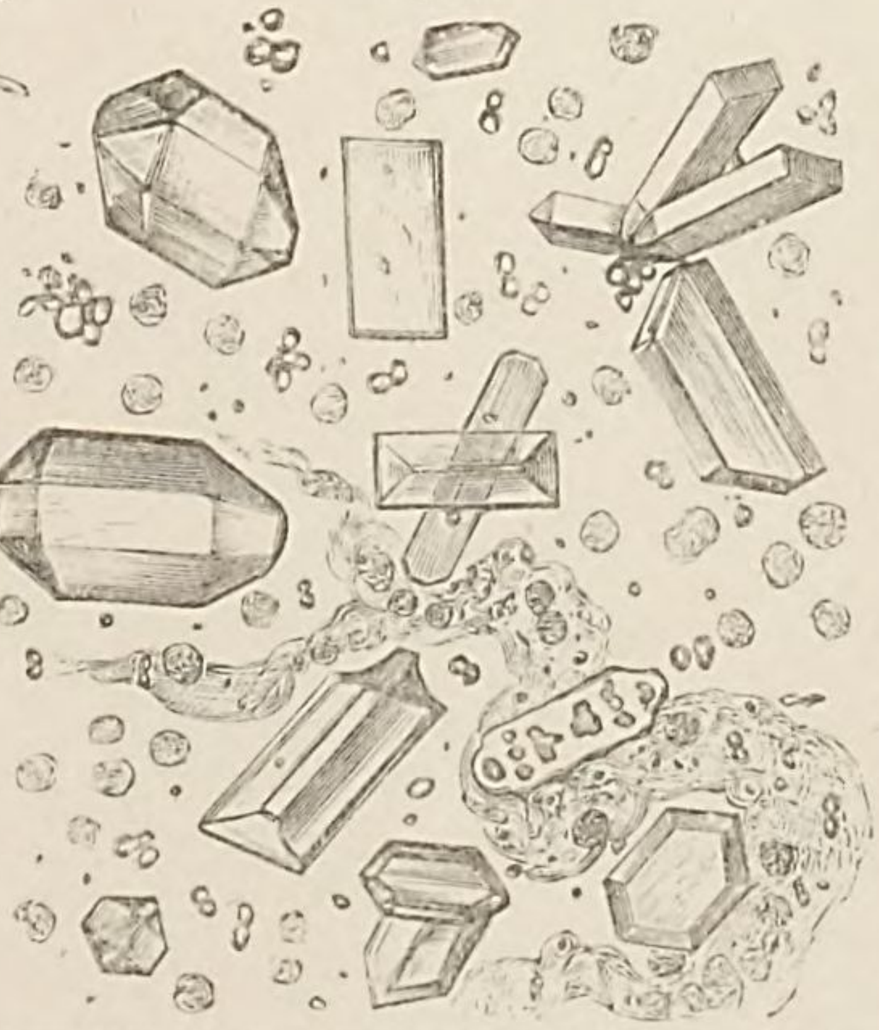
Phosphate of lime. From urine. × 130. pp. 357, 362.

Fig. 6.



Phosphate of lime. × 228. p 362.

Fig. 7.



Crystals of ammoniaco magnesian phosphate, with urates, mucus, and pus-corpuscles. From a case of catarrh of the bladder of three years standing. Man age 10. × 215. p 357.

Fig. 8.

Cystine. From urine. × 215. p. 363.

1000th of an inch ⌴ × 40.
" " ⌴ × 130.
" " ⌴ × 215.

[To follow plate LIV.

by causing solutions of phosphate of soda and chloride of calcium in strong glycerine, to mix together very gradually.

335. Cystine.—Cystine forms a deposit much resembling that of the pale urates; from which, however, it is readily distinguished by not being dissolved upon the application of heat. For the deposit from which the drawing, fig. 8, was taken, I am indebted to my friend, Dr. Sankey. An interesting case of cystine deposit, reported by Dr. Milner Barry, is given in vol. I of the "Archives of Medicine," with analyses of the urine.

Chemical Characters.—Deposits of cystine are insoluble in the warm urine or in warm water. They are dissolved by ammonia, and if the ammoniacal solution be allowed to evaporate, the six-sided crystals are again deposited. Cystine contains much sulphur, and sulphuretted hydrogen gas is among the products of its decomposition. This deposit, if incinerated on platinum-foil, leaves no fixed residue.

336. Carbonate of Lime is very rarely met with in a crystalline form in human urine. Not unfrequently it occurs, mixed with triple phosphate and phosphate of lime, as an amorphous powder, or forming very small round masses. Occasionally, however, it has been met with as dense spherical stellar aggregations of minute acicular crystals (Dr. Golding Bird). Fig. 1, pl. LIV, represents the appearance of carbonate of lime as it occurs in the urine of the horse when viewed in Canada balsam, with the aid of transmitted light, and in fig. 2, the same crystals are represented as seen by reflected light on a dark ground.

The Chemical Characters of Carbonate of Lime are described in § 203.

337. Blood Globules or Corpuscles usually form a red or brownish-red granular deposit which sinks to the bottom of the vessel. If the urine be perfectly neutral, or slightly alkaline in its reaction, the colour of the globules will be bright red; while, in those instances in which the reaction is decidedly acid, the deposit of blood will be of a dirty brown colour, imparting to the fluid a smoky hue. This smoky appearance almost always exists when the urine is acid, and the blood is derived from the kidney. In many cases in which it retains its florid colour, it has escaped from the mucous membrane of the bladder, prostate, or urethra. If blood globules remain long in urine they become much altered in form, the outline appearing irregular and ragged, and the surface granular. This character is no doubt chiefly dependent upon physical changes, pl. LVI, fig. 1.

Disintegrated Blood.—Besides blood corpuscles exhibiting various forms we not unfrequently find in certain urinary deposits the constituents of blood corpuscles in a disintegrated state. In those remarkable cases of *hæmatinuria*, quantities of altered blood are passed, but not a single whole blood corpuscle is to be detected in the urine

although it be most carefully examined many times at intervals. In these cases the blood is probably disintegrated while in the kidney before it passes into the uriniferous tubes, and, as is well known, escapes with a quantity of albumen. See note on page 372.

Chemical Characters of Urine containing Blood.—Urine containing blood corpuscles also contain serum, but the quantity of this fluid is in many cases very small, although numerous blood corpuscles are to be discovered by microscopical examination. If there be much blood, the albumen of the serum is readily detected by the ordinary reagents, but if the quantity of albumen present be greater than can be accounted for by the number of blood corpuscles, the practitioner would be led to fear the existence of organic disease of the kidney, and would at once investigate the case very carefully in order to ascertain if there was any evidence of the change. See "Casts of the Tubes," p. 353.

338. Cancer Cells.—Large Organic Globules, Inflammatory Corpuscles, Exudation Cells, &c.—Specimens of cancer cells found in the urine in cancer of the ureter and cancer of the bladder are respectively represented in pl. XLIV, fig. 1, pl. LVI, fig. 8.

The observer should be aware that in many cases the epithelial cells of the pelvis of the kidney, of the ureters, and of the bladder not unfrequently exhibit changes of form which may cause them to be mistaken for cancer cells. In the urine of a girl, aged 14, who was suffering from an attack of acute inflammation of the kidneys, I found numerous cells very closely resembling certain forms of cancer-cells, pl. LVI, figs. 9, 10. The urine contained albumen and casts with numerous blood corpuscles. The deposit was so like that of many cases of cancer and there were so many cancer-like cells that had I not known anything of the case I think I should have been led to believe that the urinary deposit came from a case of cancer of the bladder. The patient soon got perfectly well and all the suspicious elements disappeared.

Large cells filled with oil globules, which are met with in the urine in cases of fatty degeneration, have already been referred to in p. 354. These when completely filled with oil, appear perfectly dark by transmitted light and are white by reflected light. They have been termed "large organic globules," by Dr. Golding Bird, and in structure present great similarity to the so-called "exudation cells," "inflammatory globules," or "compound granular cells." They consist essentially of spherical aggregations of minute oil globules which can be readily distinguished by their dark outline and clear transparent centre. They must be distinguished from cells which are composed only of minute granules or molecules, which appear, even when examined with very high powers, as minute dots.

339. Spherical Cells containing Nuclei and Granular Matter.—

Cells exhibiting nuclei and granules are not unfrequently met with in specimens of urine, but I have not been able to determine with accuracy the portion of the mucous tract from which many of these cells have been derived, or their pathological importance. The cells represented in pl. LVI, fig. 5, were found in the urine of a patient suffering from rheumatic fever. The smaller round bodies are altered blood corpuscles. The large cells above referred to contained several transparent bodies within them, which became very distinct upon the addition of acetic acid (nuclei?). The central bodies did not refract light as oil globules, nor did they present the circular dark and well-defined outline so characteristic of them.

In pl. LVI, fig. 4, are represented specimens of large cells filled with dark granular matter, but not containing any oil particles, from the urine of a case of chronic bronchitis. There were also a few pus globules present in this specimen. It is probable that these cells consisted of altered mucus corpuscles and bodies embedded in mucus which was expectorated and afterwards thrown into the urine.

Fig. 6 represents a curious form of cell found in the urine of a case of renal dropsy of seven weeks' duration. Casts of medium diameter, with a few small cells containing oil, were also present in the same specimen of urine. Cells presenting somewhat similar characters have come under my notice in several other cases; and from that portion of the mucous surface of the bladder known as the trigone, I have obtained cells agreeing with them in general characters. It appears not unreasonable, therefore, to assume that many of these peculiar cells may result from some modification of the young and imperfectly formed cells of bladder epithelium.

Whenever a mucous membrane is inflamed, that is whenever an increased amount of pabulum gains access to the growing epithelial cells, these undergo changes which result in the production of cells differing in marked characters from normal cells. Such bodies are more quickly produced, and, therefore, the formed material of the cell is much softer than under ordinary conditions. (*See* page 247.) Oftentimes they are larger and they are generally spherical, or oval, and, therefore, contrast remarkably in form with such forms of epithelium as the scaly, tesselated, and columnar, though developed from masses of bioplasm, which in the ordinary course would have produced these.

340. "Small Organic Globules."—Under this name Dr. Golding Bird has described some little bodies smaller than the pus or mucus corpuscles, with a perfectly smooth exterior, and unaffected by acetic acid. Dr. Bird suggests that they may be nuclei which have been set free from a cell by the bursting of the investing membrane, but this is not very probable. Fig. 11, pl. LVI, represents the appearance of the deposit from the urine of a patient suffering from calculus. The small

round bodies represented in different parts of the figure were insoluble in strong acetic acid, and were unaltered on the addition of ether or potash. Many of them contained a central dark spot. They were accompanied with numerous small octahedral crystals of oxalate of lime. From their highly refractive properties and chemical characters just referred to, it is most likely that they were composed of oxalate of lime.

There are other small round bodies met with from time to time in urinary deposits, the nature of which it is not easy to determine. Some of these consist of altered blood corpuscles, others are sporules of fungi, fig. 7, pl. LVI, while small spherical crystals of oxalate of lime are sometimes present.

It is most desirable that when the practitioner meets with objects, the nature of which he cannot ascertain, careful drawings should be made, §§ 76 *et seq.*, and the specimen preserved if possible. Notes of the case should also be carefully kept. Of course care should be taken that the observer is not misled by the appearances of the extraneous substances likely to be met with, p. 344, and he should therefore make himself familiar with these as soon as possible.

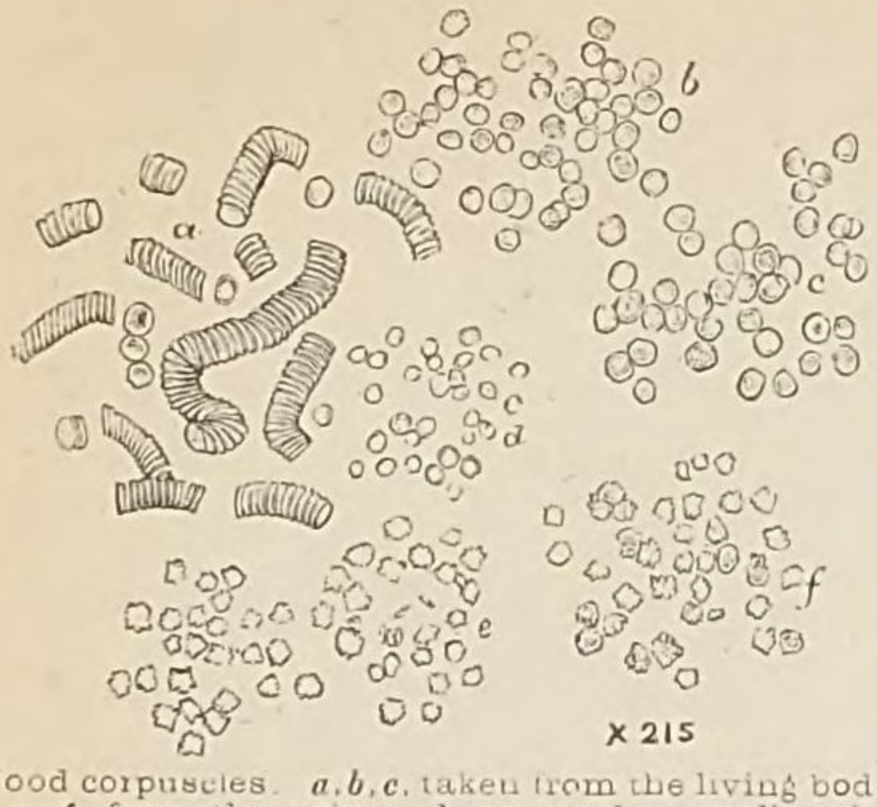
Urinary Calculi.

341. Formation of Urinary Calculi.—The structure and formation of urinary calculi have been studied with the aid of the microscope, and most important facts have been recently ascertained concerning their origin. It is important to bear in mind that there was a time when even the largest calculus was a microscopic object,—when it might have been removed from the organism, and the formation of 'stone' entirely prevented.

I have found that the 'nucleus' of almost all calculi possesses the same composition and exhibits the same characters. If small *uric acid* calculi be soaked in liquor potassæ, the uric acid will be dissolved, and a 'nucleus' will be left which consists of a little mucus with small fragments of oxalate of lime; and in very many cases *well-formed dumb-bell crystals* of oxalate of lime may be clearly demonstrated. The same remarks apply to other calculi, and of course to the ordinary oxalate of lime calculi themselves. And I have been gradually led to the conclusion that *dumb-bell* and *spherical crystals of oxalate of lime*, which I have shown are formed in the uriniferous tubes, very frequently constitute the 'nuclei' around which other insoluble or partially soluble materials are deposited. The mode of formation of the nuclei has been studied in specimens washed away from the kidney, and in some found in the uriniferous tubes of kidneys obtained from the post-mortem room.

These interesting facts with regard to the formation of the nucleus

Fig. 1.

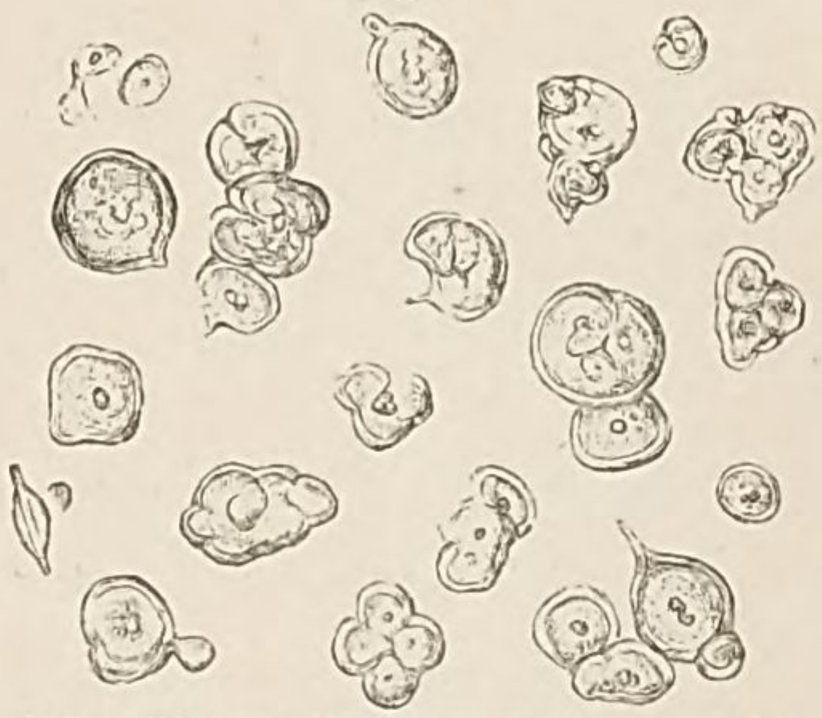


Blood corpuscles. *a*, *b*, *c*, taken from the living body; *d*, *e*, *f*, from the urine. *d*, corpuscles smaller than natural; at *e* their circumference is serrate and ragged; and at *f* a somewhat similar appearance is shown. p. 363.

Fig. 2.

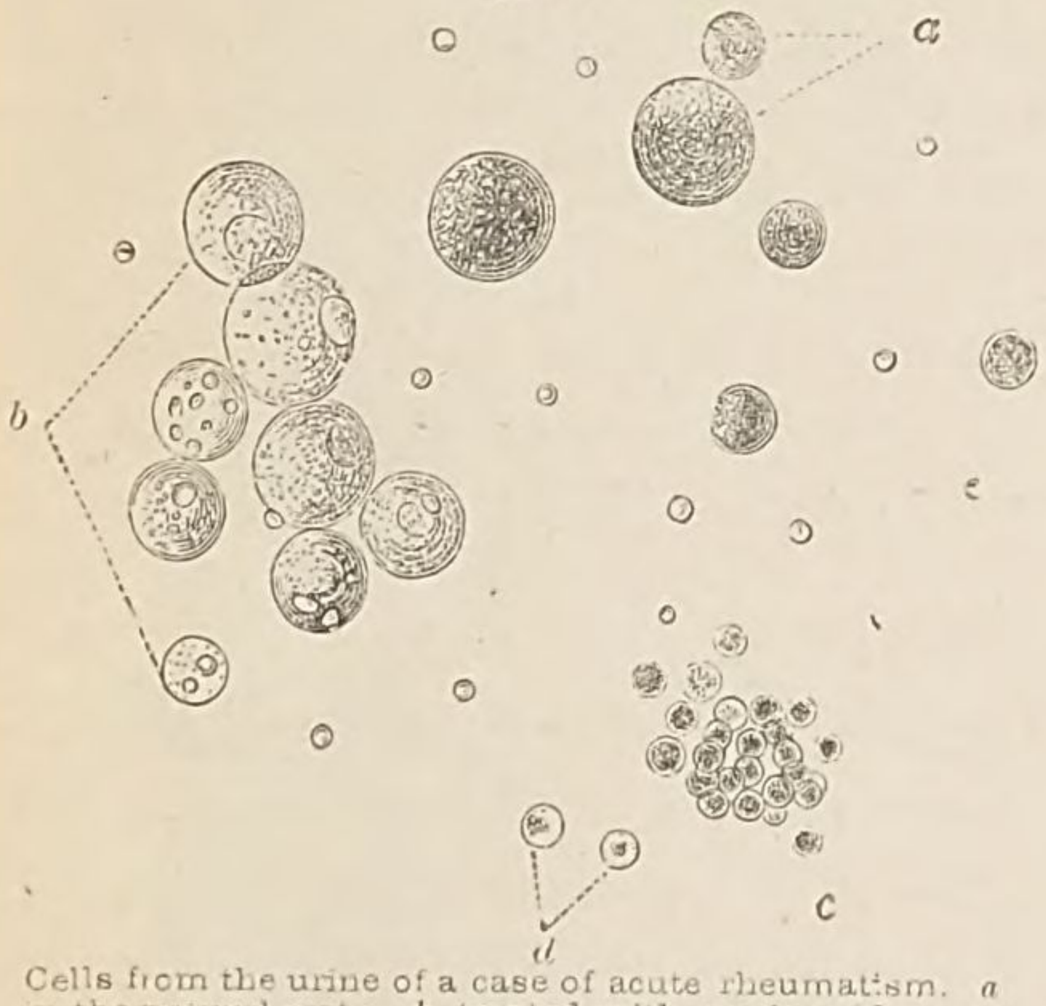
Blood corpuscles with spots; from the urine of a case of calculus in the kidney. Almost every corpuscle exhibited a little round spot like that displayed when blood corpuscles are treated with a solution of aniline. The crystals are crystals of hæmatoidin. × 215. p. 372.

Fig. 3.



Some of the corpuscles represented in Fig 2. × 700. p. 372.

Fig. 5.



Cells from the urine of a case of acute rheumatism. *a* in the natural state. *b*, treated with acetic acid *c*, resembling pus *d* the same treated with acetic acid. The small circular bodies are blood corpuscles. p. 365.

Fig. 8.

Cancer cells found in urine. From the bladder p. 361.

Fig. 4.

Large cells from the urine of a case of chronic bronchitis. p 365.

Fig. 6.

Cells found in the urine of a case of renal dropsy. p. 365.

Fig. 7.

Sporules of fungi, resembling blood corpuscles. p 366.

Fig. 10.

From a case of acute inflammation of kidney. Girl age 14 Showing altered cells of renal epithelium with modified cells of bladder epithelium, many of them very like cancer cells. Blood corpuscles and pus-like cells are also present. × 215. p. 361

Fig. 11.

Small globules and octahedra of oxalate of lime. p 365.

Fig. 9.

An altered cell of bladder epithelium, showing the mode of formation of pus within the cell. From a case of irritation of the bladder. × 215. p. 364.

1000th of an inch ⌊—⌋ × 215.

" " ⌊————⌋ × 700.

[To face page 366

of an urinary calculus have not been noticed or referred to by Dr. Vandyke Carter in his book published in 1873. (The Microscopic Structure and Mode of Formation of Urinary Calculi, 1873.) Indeed, it would appear from several remarks in that volume, which are quite confirmatory of my observations, that Dr. Carter was not at the time aware that I had written upon the subject. In his preface, however, he gives references and makes acknowledgments which ought to have been inserted in the text. As it is the reader would infer from Dr. Carter's figures and observations that certain facts were first made out in 1873, while in fact they were known before and were published in the year 1861 in the first edition of my work on urine, &c. Indeed, I was studying the formation of microscopic calculi as far back as 1855, although Dr. Carter says in 1873, that the structural composition of calculi "has hitherto been but little noticed." He does not seem to be aware of the great frequency of dumb-bell crystals of oxalate of lime in the nucleus of uric acid calculi. Neither does he appear to take cognisance of the facts proving that these crystals are found in the tubes of the kidney—a matter as already shown of considerable importance in connection with the origin of urinary calculi.

Fig. 6, pl. LIV, represents a mass of dumb-bell crystals. Many such collections were passed by the same person, and I have met with similar collections in several different individuals. Although the mass is seen to consist of a number of distinct crystals, these latter are firmly attached to each other, so that the whole may be rolled over and over while under observation without the individual crystals being separated from one another. Such collections I have many times seen in the uriniferous tubes of the kidney; a fact which indisputably establishes the precise seat of formation of these bodies. As time goes on the interstices between the individual crystals gradually become filled up with a similar material, and at the same time a few of the larger crystals increase in size at the expense of the small ones. At length a small crystalline mass of an oval form results, which is in fact a microscopic mulberry calculus. If this had remained in the uriniferous tubes, it would have gradually increased in size. Two small calculi of this description are represented in my work on "Kidney Diseases, Urinary Deposits, and Calculous Disorders." When such calculi reach the bladder, they doubtless gradually increase by the deposition of various salts upon their surface. Such small bodies might easily become entangled in the mucus, and might remain in the pelvis of the kidney without exciting any disturbance until they had grown as large as a pea or still larger, when great inconvenience and perhaps very severe pain might result.

The above observation is of interest also as showing the chemical composition of the dumb-bells, which has long been a disputed point.

It is very difficult to obtain sufficient of the deposit of the dumb-bell crystals for an accurate chemical examination, but we know that the mulberry calculus consists of oxalate of lime, and as I have shown that it is composed of aggregations of dumb-bell crystals, there can be little doubt concerning chemical composition of an individual dumb-bell crystal. It is of importance that cases in which these dumb-bell crystals are deposited should be very carefully watched and the secretion of plenty of urine encouraged, so that the dumb-bells may be washed out of the tubes as soon as possible.

Microscopic calculi, composed of mucus, altered epithelium, and phosphate of lime, are also formed in the prostate gland, and several specimens are represented in "Kidney Diseases, Urinary Deposits, and Calculous Disorders."

A microscopical calculus having been formed, the same material may be added upon its surface, layer after layer, or if the urine happen to be rich in uric acid, urates, or phosphate, layers of these salts will be deposited. As is well known, it often happens that from time to time the characters of the urine become modified, and layers of different salts are deposited. On section the calculus is seen to consist of concentric layers.

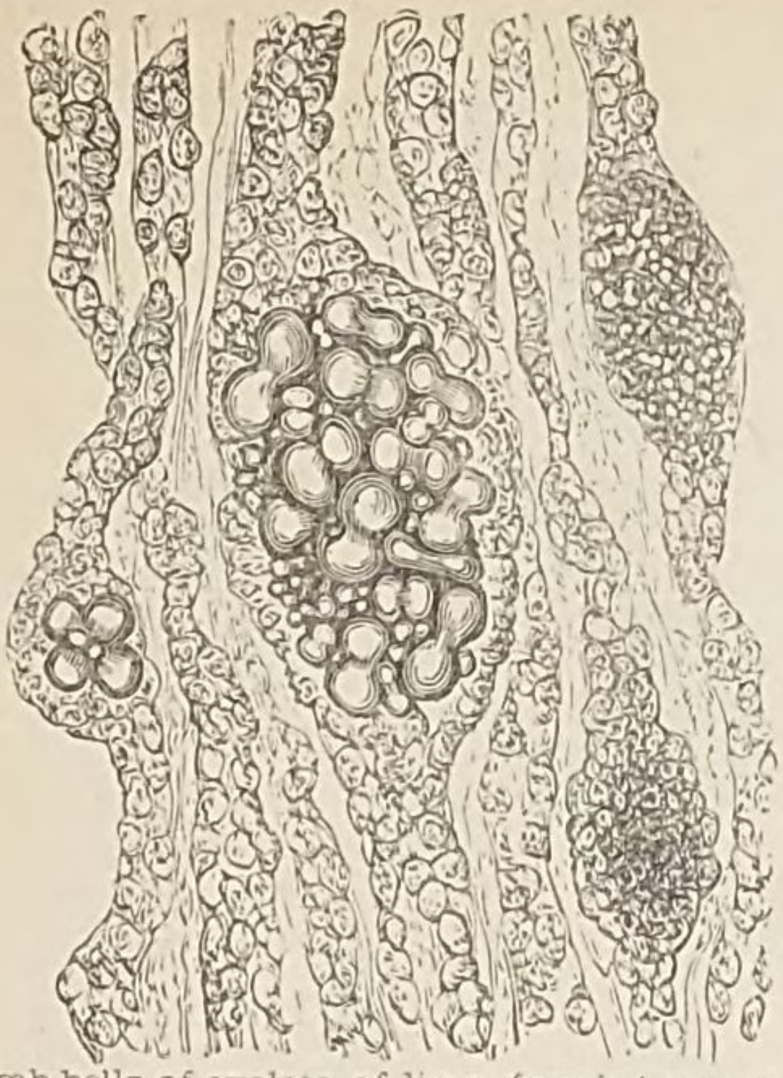
The physician has two objects to attain in the management of the health of persons who are the subjects of calculous disorders—

1. To expel from the kidney the dumb-bells which may form the 'nuclei' of calculi, and to prevent the formation of dumb-bells and spherules of oxalate of lime.
2. To prevent, as far as possible, the development of the various conditions of urine which favour the chemical changes which lead to the deposition of uric acid, urates, and phosphates.

342. Of Fragments of Calculi.—Small portions of calculi, especially of the softer forms of the triple or ammoniaco-magnesian phosphate calculus are often passed in the urine by those who are troubled with calculus. By microscopical examination with an inch or two-inch object glass, or even by the use of a still lower power, the nature of the particle may generally be determined. Should the observer, however, be in doubt, a small portion of the fragment must be detached, crushed upon a glass slide and tested according to the plan described in page 169; the action of the test being carefully studied with the aid of the microscope.

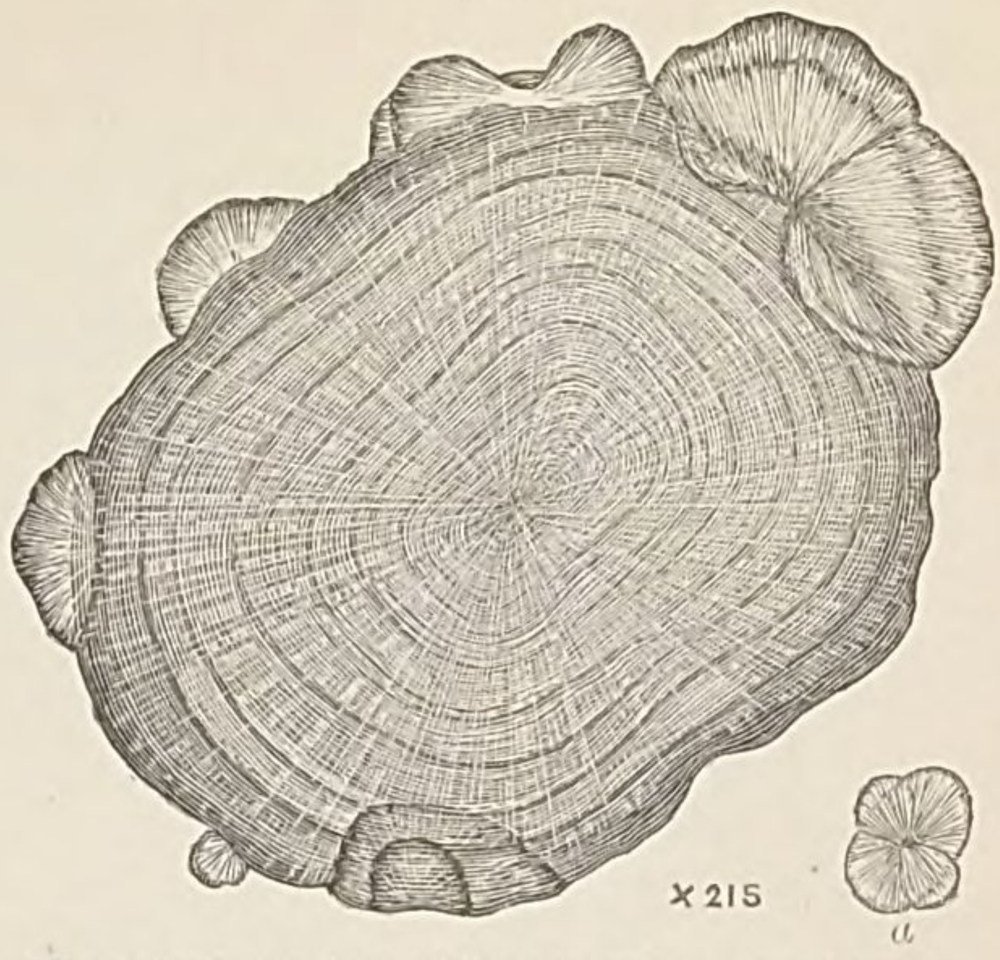
The fracture of calculi within the bladder and even while they remain in the pelvis of the kidney undoubtedly occurs, apparently without violence—certainly in the absence of the degree of pressure or concussion that would be necessary to effect the same fracture out of the body. One can conceive that when several stones are together in a cavity a very sudden movement of the body might cause them to knock

Fig. 1.



Dumb bells of oxalate of lime, forming minute calculi, impacted in the uriniferous tubes of the kidney. × 215. p. 367

Fig. 2.

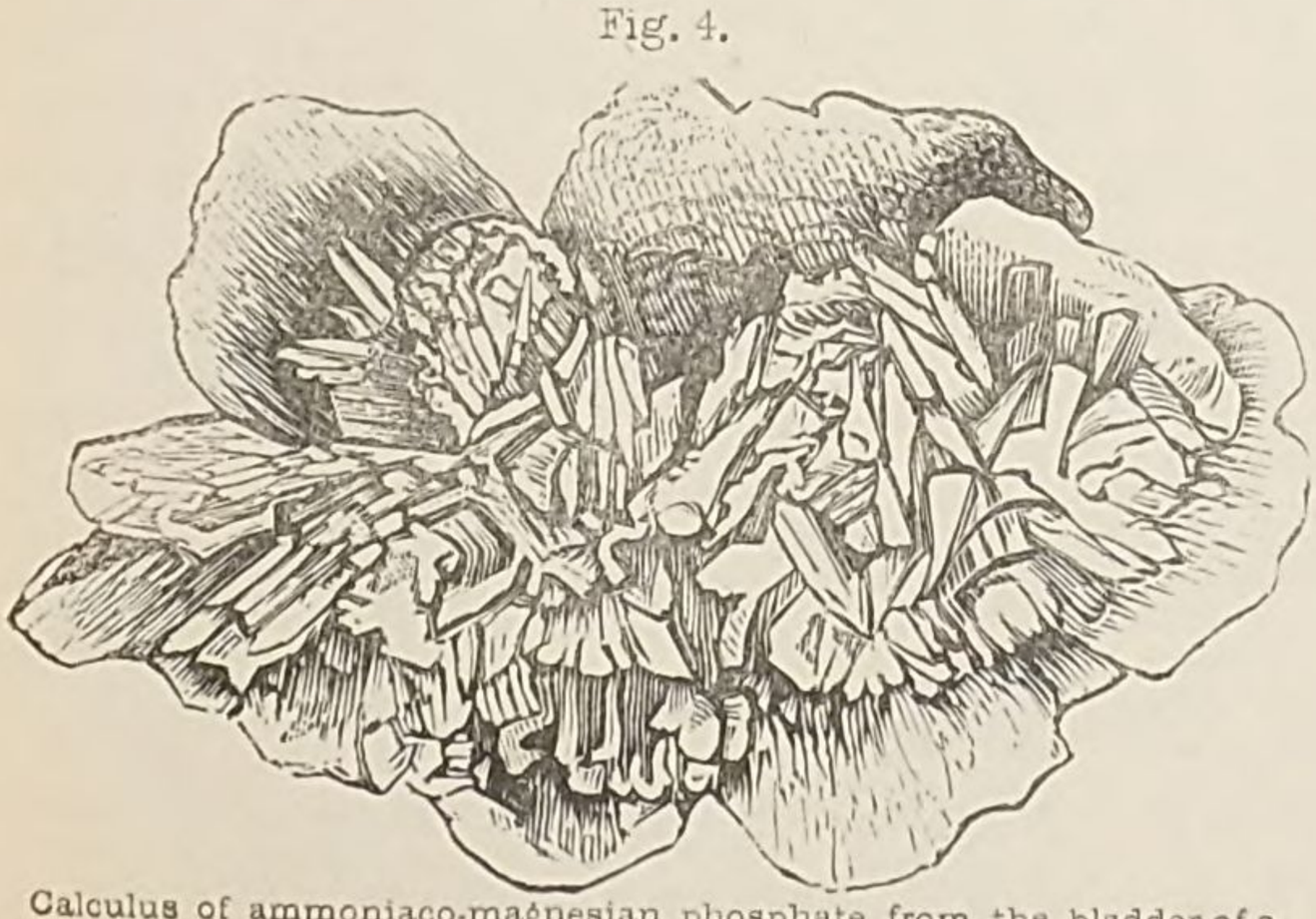


Microscopic oxalate of lime calculus passed in the urine, consisting of dumb-bells of oxalate of lime and one very large oval mass × 215. p. 367

Fig. 3.

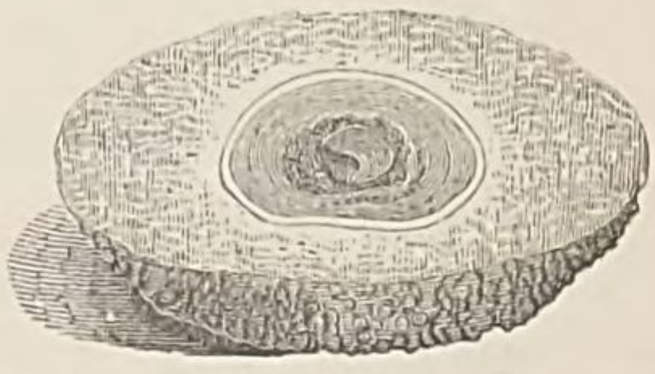
Microscopic calculi composed of oxalate of lime, from a patient suffering from symptoms of renal calculus × 215. p 367.

Fig. 4.



Calculus of ammoniaco-magnesian phosphate from the bladder of a dog. × 5. Messrs Gowing & Son.

Fig. 5.



Phosphatic calculus, the nucleus being composed of uric acid.

[To face page 368

1/000th of an Inch × 215.

Fig. 1.

Blood calculus from one of the infundibula of a kidney.

Fig. 2.

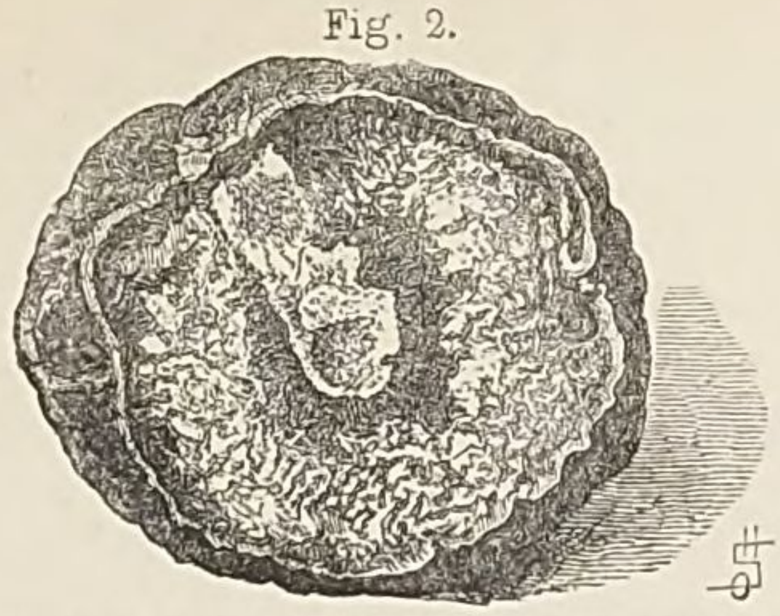
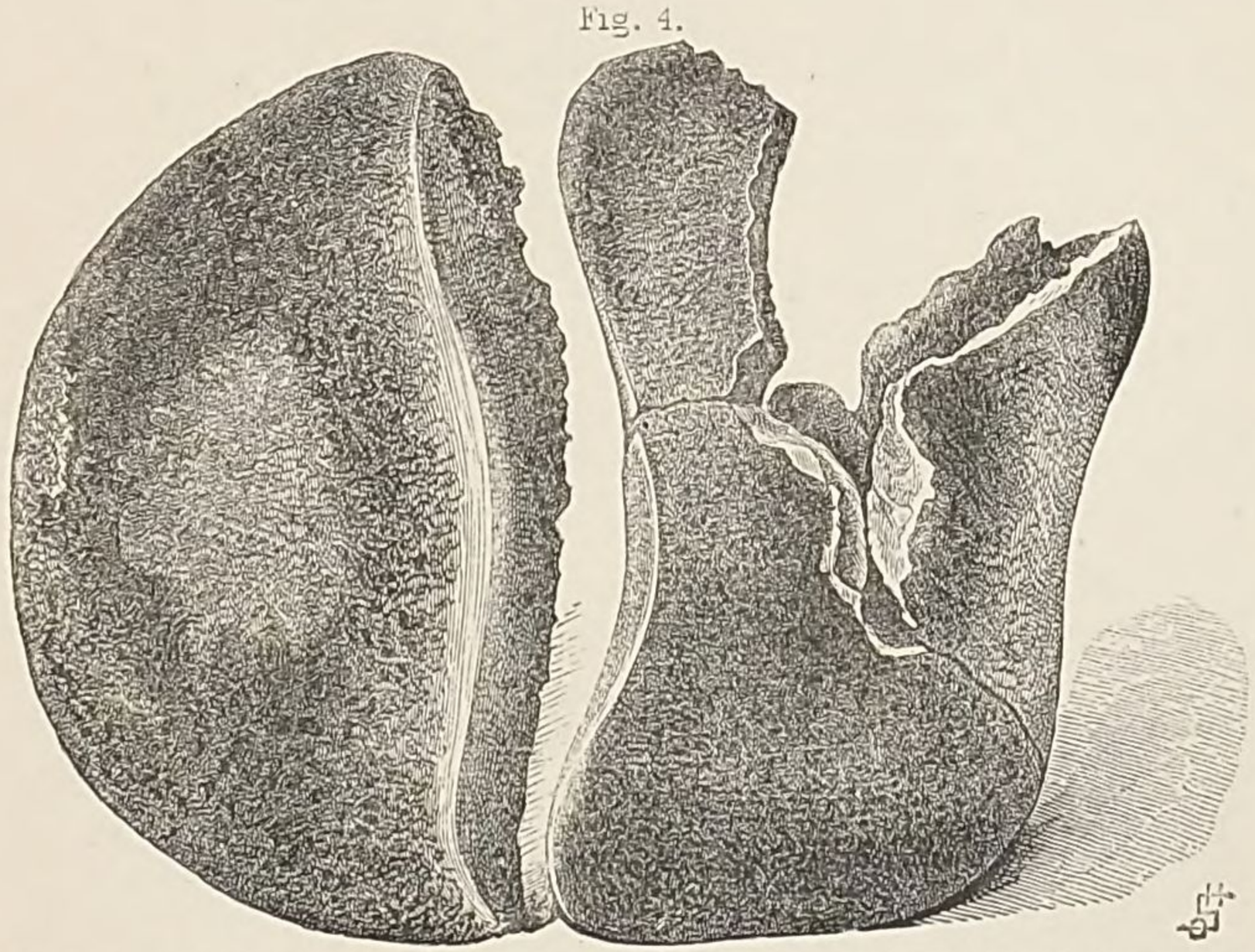


Fig. 3.

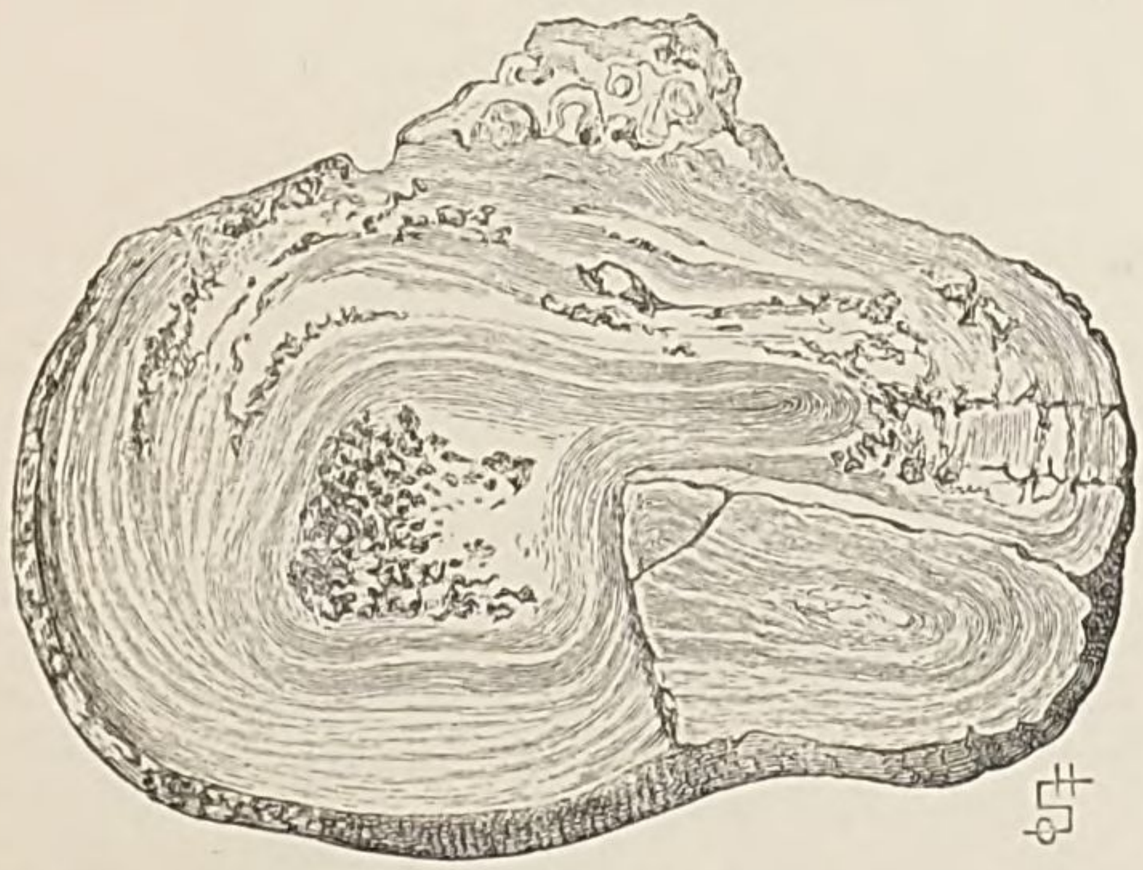
Case of spontaneous fracture of a calculus in the bladder. Drawn by Mr. Southam. p. 369.

Fig. 4.



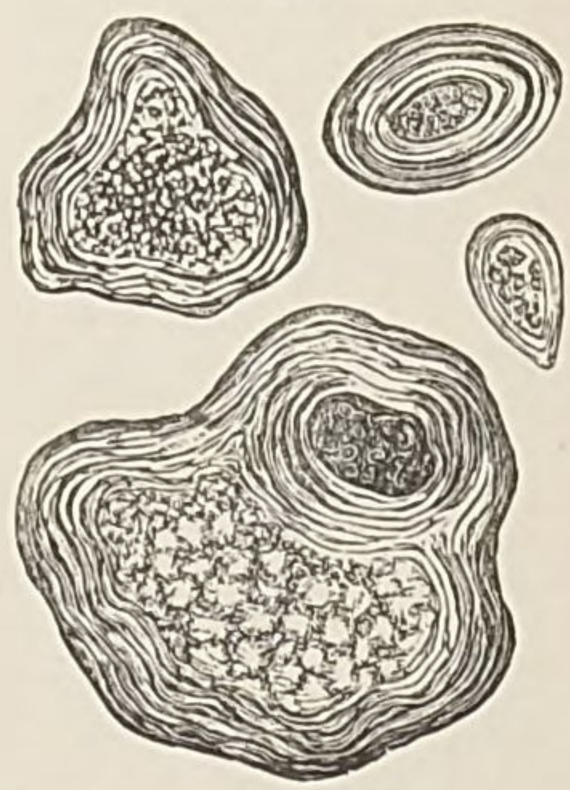
Spontaneous fracture of a very large stone in the bladder. The case was operated on by Mr. Luke. Drawn by Mr. Southam. p. 369.

Fig. 5.



Spontaneous fracture of a calculus in the bladder From the Dupuytren Museum. Drawn by Mr Southam, p. 369. The separated portion seems to have become again cemented, and encrusted with subsequent deposit.

Fig. 6.



Small phosphatic calculi from the kidney. Nuclei consisting of epithelium. × 130.

[To follow Plate LVII.

against one another violently enough to fracture the softest of them, or a soft stone might be broken by the impaction of the sound or catheter introduced when the bladder was examined. But spontaneous fracture of a single stone has taken place in several instances—even when the stone was composed of matters as hard as uric acid and oxalate of lime, and when there had been no sudden shake of any kind. In such cases it is probable that some decomposition takes place in the substance of the calculus itself, and that the pent up gases gradually exert sufficient pressure to break up the mass. Layers are sometimes stripped off from this cause, as in the case figured by Mr. Southam from the Dupuytren Museum, Paris. *See* pl. LVIII, fig. 5.

Mr. Southam reports two cases of spontaneous fracture of calculi in the bladder. (Brit. Med. Journ., July 4th, 1868.) The first occurred in a boy of 15 and the calculus was uric acid. The stone and the fragment broken off from it spontaneously are represented in figs. 2 and 3, pl. LVIII. The second case occurred in a boy aged 7. The fracture had occurred in the bladder probably many months before the boy was operated upon. Mr. Southam also reports a case operated on by Mr. Luke—in which a very large stone, fig. 4, pl. LVIII, was removed from the bladder of a man aged 63. It was found broken into the two very large fragments represented in the drawing.

On the Preservation of Urinary Deposits.

It is very desirable to permanently preserve many urinary deposits, particularly when their nature is doubtful, in order that they may be compared with other specimens. As the student will meet with many difficulties in his attempts to preserve the characters of urinary deposits, it will be advantageous to consider the subject somewhat in detail. While some of the substances met with may be easily preserved, others are only prevented from decomposing by taking very great care in mounting them, and by the use of good preservative solutions. There are three methods of mounting urinary deposits in ordinary use:—1st. As dry preparations. 2nd. In Canada balsam, turpentine, oil, and other fluids of similar characters. 3rd. In an aqueous preservative solution.

The first method is only applicable in a very few cases, as the greater number of substances forming urinary deposits are so altered by the processes of washing and drying as to be afterwards recognized with difficulty. Large crystals of uric acid, crystals of oxalate of lime, and certain forms of phosphates and urates may, however, be mounted as dry objects, but they of course exhibit different appearances when examined in fluid.

343. Preservation of Urinary Deposits in the Dry Way.—Specimens which are to be mounted in the dry way must undergo the same

preliminary washing and drying as those which are to be put up in Canada balsam. The same course of procedure, therefore, applies to both cases. Suppose we require to dry some crystals of uric acid:—after the crystals have been allowed to collect at the bottom of a conical glass vessel, the clear supernatant fluid is to be poured off, and the crystals are to be washed with a little dilute alcohol, or with a very weak solution of acetic acid. When the process of washing has been repeated two or three times, a small quantity of the deposit is to be transferred by means of a pipette to a glass slide, and the greater part of the fluid soaked up with a small piece of blotting-paper. The crystals are next to be spread a little over the glass, with the aid of a fine needle, in order to separate the individual crystals from one another, and the slide is to be placed in a warm place, or in the sun, until quite dry; but care must be taken that the drying is not carried on too rapidly, and that too great a degree of heat is not employed. A narrow rim of paper or cardboard is next to be gummed on the slide so as to include the crystals in a sort of shallow cell; and, lastly, the glass cover is to be put on, and kept in its place either by anointing the edges with a little gum water, or by pasting it down with narrow strips of paper, which may be variously arranged and ornamented according to taste.

344. Preservation of Urinary Deposits in Canada Balsam.—If the crystals of uric acid are to be mounted in Canada balsam, they should be carefully dried first, as above directed, and afterwards placed over sulphuric acid under a bell jar. When quite dry the crystals are to be moistened with a small drop of spirits of turpentine. The slide is then to be slightly warmed, in order to volatilize the greater part of the turpentine, and a drop of Canada balsam is to be dropped upon the preparation from the end of a wire. This may be readily effected by holding the wire with the balsam over the lamp or hot brass plate for a few seconds, in order to soften it. The slide is next to be held over a lamp, or placed upon a hot brass plate, in order to keep the balsam fluid until any air bubbles which may be present have collected into one spot on the surface of the liquid balsam, an operation which is expedited by gently moving the slide from side to side. The air bubbles may now be removed by touching them with a finely-pointed wire, a needle or pin. Lastly, the glass cover is to be taken up with a pair of forceps, slightly warmed over a lamp, and one edge allowed to touch the balsam. The glass is permitted to gradually fall upon the balsam, so that it may be wetted by it regularly, and only by slow degrees. In this way we may prevent air bubbles from being included in the preparation. The glass slide may now be set aside to cool.

345. Preservation of Urinary Deposits in Aqueous Solutions.—For the preservation of urinary deposits, the most important method is to

keep them in some preservative fluid, miscible with water, for in this way only can the characteristic appearance of many specimens be retained. Mounted in the dry way, and in Canada balsam, it need scarcely be said that the object presents different characters to those demonstrated when it was immersed in the urine; and although those methods are of advantage in demonstrating the internal structure or arrangement of some crystals they are ill adapted for the preservation of the great majority of urinary deposits, while for the preservation of epithelium, casts of the renal tubes, &c., they are wholly inapplicable.

When preservative solutions are employed, the objects must always be placed in shallow cells; and the most convenient form of cell for this purpose, according to my experience, is that which is made by painting upon the glass slide, with a fine brush, a narrow border of Brunswick black, inclosing either a square or circular space as may be most convenient. In cases where a deeper cell is required, those composed of thin glass or tinfoil are the most useful. The forms of cell just referred to, I can recommend from experience, for I have many preparations put up in them which have been well preserved for more than twenty years. H. to W. §§ 116, 117, 118.

Preservative Solutions.—Next, with regard to the preservative fluids best adapted for mounting urinary deposits;—weak spirit answers pretty well for some sediments, but as a general rule is not suitable for substances found in urine. Glycerine may be employed in many cases diluted with a little water or spirit. The preservative gelatine I have found answer exceedingly well for the preservation of dumb-bell crystals of oxalate of lime and some other crystalline deposits: with care, epithelium may also be preserved in it. I have used the creosote and naphtha solution most successfully for the preservation of casts and various kinds of epithelium, &c., pp. 54, 55.

Whatever preservative fluid is used, care should be taken that the deposit to be put up is thoroughly saturated with it before it is mounted, for unless this object be attained, there is danger of the preparation deteriorating before many months have passed. Glycerine if used must be added to the deposit in the conical glass in very small quantities at a time, so that cells, casts, and other soft bodies may swell out again after they have been caused to shrink.

346. Method of Separating the Deposit from the Urine, and placing it in the Preservative Fluid.—The most simple manner of mounting deposits in aqueous preservative fluids is to allow the sediment to subside to the bottom of a conical glass, the supernatant urine being poured off, and a small quantity of the preservative solution added. The deposit is again allowed to subside, and the solution poured off, and replaced by a fresh quantity. This method must be followed when glycerine, the naphtha and creosote fluid, or carbolic

acid water is selected as the preservative fluid. After the subsidence of the deposit, a small portion may be removed with a pipette, placed in one of the forms of cells above referred to, and the glass cover applied to the surface of the liquid, care being taken that the whole surface of the glass is well wetted with the solution, in order that no air bubbles may be included in the preparation. Any excess of fluid is now to be soaked up with a clean cloth, or with blotting paper, and the cover cemented to the cell by applying a little Brunswick black, Dammar, or other varnish with a camel's hair brush. The name of the deposit, with any other particulars, is to be appended to the slide, and the preparation laid flat in the cabinet.

In the manner just detailed, the following may be readily preserved: various kinds of epithelium, casts, oil-globules, torulæ, confervæ, pus, mucus, uric acid, oxalate of lime, urates of soda and ammonia, and other substances, whose characteristic appearance is not altered by aqueous fluids. If uric acid, oxalates, phosphates, or other crystals, are to be put up as objects for examination with polarised light, they should be mounted in strong glycerine, balsam, or turpentine.

347. Preservation of Crystals of Triple Phosphate. Cystine.—Crystals of the triple phosphate may be preserved in water to which a little ammonia and muriate of ammonia have been added. In this solution the crystals preserve their beautiful smooth character, while in pure water or in creosote fluid the surface becomes roughened. Dumb-bells, as I before noticed, may be preserved in the preservative gelatine, and they are not liable to shift their position in consequence of being well supported by the jelly. Crystals of cystine cannot be preserved in the creosote solution, because they are slowly dissolved by it; but as they are insoluble in organic acids, a very weak solution of acetic acid will keep them unchanged. If cystine or uric acid is to be mounted in glycerine the fluid should be made acid by the addition of a very little acetic acid.

Note to § 337, p. 363.—Some blood corpuscles exhibiting unusual characters are represented in pl. LVI, figs. 2 and 3. Each exhibits very distinctly a roundish dot, exactly resembling that which is seen when blood is treated with a solution of aniline, as was first demonstrated by Dr. Roberts. In fig. 3 the contents of the corpuscles have shrunk so as to leave a space between them and the outer hardened part of the corpuscle often spoken of as the "cell wall." The little spot is probably the remains of the bioplasm from which each corpuscle was developed.

CHAPTER XVII.

OF EXAMINING VARIOUS TISSUES AND ORGANS IN HEALTH AND DISEASE.—*Areolar Tissue.—Adipose Tissue.—Cartilage.—Osseous Tumours and Plates.—Myeloid.—Striped Muscle.—Muscular Fibre in a state of Fatty Degeneration.—Examination of Unstriped Muscle.—Examination of Nerve-Fibres.* MICROSCOPICAL EXAMINATION OF ORGANS.—HEART.—*Arteries and Veins.—Small Arteries.—Capillaries.—Atheromatous and Calcareous Deposits in Arteries.* LUNGS.—*Trachea and Bronchial Tubes.—Pulmonary Tissue in Pneumonia, in Tubercular Disease, in Cancer.* ALIMENTARY CANAL.—*Of Mucous Membrane generally.—Epithelium.—Submucous Tissue.—Tongue, Salivary Glands, and Parts about the Mouth.—Thickening of the Submucous Tissue of the Stomach and Intestines.—Ulcers of the Intestines.*

THE general method of investigating the structure of tissues has been already placed before the reader, and in Chapter VII (page 99) I have described the process which in my hands has afforded most satisfactory results. The principles laid down and the method of proceeding described will be found applicable to almost any tissue or organ the structure of which it may be desired to investigate. The plan of staining by injecting the vessels may be carried out in the case of any textures of the body in health or in a morbid state and is equally applicable to morbid growths. But, since in the study of certain organs special methods have to be resorted to, it may be well to recount some of the most important of these under their proper headings in the present and succeeding chapters.

For investigating the arrangement of the minute anatomical elements of some tissues and for determining the general order of growth and the manner in which the structural elements have, so to say, been laid down, advantage may be gained by employing certain plans in conjunction with the particular method of proceeding which I have advocated. For general investigation, it has been already pointed out that nitrate of silver, chloride of gold, osmic acid, and some other substances used in the preparation of tissues for examination with very high powers, may be dissolved in glycerine, and the sections of tissues

subjected to the action of such solutions (the strength of the solution and the time of soaking being varied according to the effects it is desired to produce) preserved in it. Lines, fibres, inequalities of surface, delicate gradations in the density and transparency of a texture may thus be demonstrated, of which not a faint indication could be discovered in the tissue when submitted to examination in the usual manner. In many instances the observer will be surprised at the very minute quantity of the substance that will afford him good results. A solution containing less than half a per cent. of solution of chloride of gold is strong enough to bring out many points not previously observable. Too much caution cannot, however, be exercised as regards the interpretation put upon the new appearances produced. Want of care and patient consideration before strongly committing himself to a definite opinion has led many an observer into very serious mistakes, and, what is worse, has caused him unwittingly to increase that most painful of all labours connected with the progress of science, the labour of controverting erroneous observations.

348. Areolar or Connective Tissue is the seat of many important changes in disease. The connective tissue bioplasts are always enlarged in inflammation and fever, and the two kinds of formed material, the white and the yellow fibrous tissue, are more abundant, and the fibres thickened in many forms of skin disease. Areolar tissue can always be obtained from beneath the skin, and mucous membranes, or from the external coat of the arteries. In some situations it is lax and very abundant. It may be blown up with air, and dried to show the areolæ or spaces in which it is disposed. If the vessels be injected with plain size, the areolæ become distended with it, and when the tissue is cold, very thin sections may be easily cut which show the arrangement of the fibres in the most beautiful manner. It is frequently associated with adipose tissue, but beneath the skin of the eyelids and scrotum, and in some other situations, it exists free from fat.

Many tumours and morbid growths are composed of a modification of areolar tissue. Both the white and yellow element, are, however, in most cases coarser than the same structures in healthy areolar tissue. Such growths are often very firm and unyielding, with few vessels distributed to them. The investigation is conducted in the same manner as that of healthy textures. In pl. LIX, figs. 1 and 4, specimens of the white fibrous element are represented. An example is figured in pl. LIX, fig. 2, which represents modified yellow elastic tissue which formed the chief constituent of a very large scrotal tumour removed by Sir William Fergusson. Some fibres of healthy yellow elastic tissue are also seen in fig. 5 in the same plate.

349. Adipose Tissue.—Many tumours are composed entirely of a tissue which could not be distinguished from normal adipose tissue, but

in some the adipose vesicles are much larger than they are in the healthy tissue. *See* pl. LIX, figs. 3, 6.

The mesentery of the mouse is very advantageous for the study of the structure and growth of this texture, which should be examined by low as well as by high powers (a two-inch, or an inch, and a quarter of an inch object-glass), and by reflected as well as by transmitted light. The bioplasm may be demonstrated by staining with carmine, and most instructive specimens may be obtained from the young frog or newt, or from the mesentery of any young animal.

Fat cells are not unfrequently found degenerated in emaciated subjects, the fat having much diminished in quantity, and the greater part of the cell being occupied with a serous fluid, which, however, exists in very small proportion in a state of health. Under these circumstances the bioplasm is often very distinct. The cell may also be in a shrivelled condition, and of a more irregular or angular form. The fluid in its interior has been found to contain granular matter with many small oil globules.

350. Cartilage.—The general characters of cartilage are very easily demonstrated. A thin section may be placed in water or glycerine. Specimens should be taken from the larynx, rings of the trachea, the ear, the ribs, the articular cartilage of joints, and the fibro-cartilage between the vertebræ and in other situations. The ear of the mouse perhaps affords the best example of cartilage consisting almost entirely of cells. The thin part in the upper extremity of this cartilage is very favourable for studying the nutrition and mode of growth of the little cells. The so-called intercellular substance or matrix is very small in quantity in many varieties of membraniform cartilage, and in some hardly exists. Specimens of cartilage keep very well in dilute spirit and water, creosote fluid, and many other solutions, but upon the whole I prefer glycerine as a medium for their preservation.

Many peculiar markings and fibrous-like arrangements have been seen in the so-called matrix of different forms of cartilage. According to many, these markings indicate a definite structure, but it seems to me, judging from the drawings of others and from the actual appearances I have myself observed, that the lines and differences in tint are probably due to the different degrees of tension, density, and moisture of the matrix around each bioplast, and not to any structural peculiarity. Like every other texture in the body cartilage is formed gradually and interruptedly, and the cartilage material is deposited at a varying rate. No wonder then that an appearance in one place of a lamellar, in another of a fibrous character of the matrix should exist. No wonder that, as the process of drying and contraction under varying degrees of stretching or pressure goes on, spaces, rents, fissures of the most varied character should be developed. When all these inequalities of texture

with the necessarily accompanying deficiencies of permeability are rendered evident by chloride of gold, nitrate of silver, or other salt capable of being decomposed by the action of light, very striking and remarkable arrangements of outline, light, and shade are rendered manifest, but that these appearances are really due to any constant structural characteristics of the tissue would seem to be at least in the present state of our knowledge an unjustifiable proposition.

In preparing sections of diseased articular cartilage, a strong knife should be used, in order that a small portion of the subjacent bone may be obtained in connection with the perpendicular section of the cartilage. In disease, fat globules are often found in considerable number in the cells of articular and other forms of cartilage. In certain cases of fatty degeneration of the margin of the cornea, *arcus senilis*, the change is associated with fatty degeneration of the muscles of the cartilages of the ribs and larynx and other tissues, as was first demonstrated by my friend Mr. Canton, of Charing Cross Hospital.

Morbid growths, closely resembling ordinary cartilage in structure, were first described by Müller under the term of *enchondroma*. They are most frequently connected with the bones of the extremities, but are occasionally developed in glands and soft parts. In their general characters and minute structure they closely resemble cartilage, but the arrangement and number of the cells vary greatly in different specimens of the morbid growth.

In the *pulpy degeneration* of cartilage, the softening is due to an increased growth of the bioplasm of the so-called cartilage cells, and to alterations in the matrix consequent upon this change, and the passage of far greater quantities of nutrient fluid through the tissue than in the healthy state. Professor Goodsir was, I believe, the first to show that the cells (bioplasm) were augmented considerably in size, and gave origin to a number of smaller ones (bioplasts, new centres) in their interior. The different stages through which these bodies pass in their development, have been well described by Redfern. "On Anormal Nutrition in the Human Articular Cartilages," 1852.

351. Osseous Tumours—Bone-like Structures.—The general method of examining bone and the manner of making thin sections have been already described in Chapter VII, p. 108. Sections of bony tumours, which have the structure of true bone, and of the hard plates, such as those frequently found in the coats of arteries, which are merely masses of calcareous matter deposited in a lamellar form, may be obtained in the same way as sections of bone. Good sections of tumours which have partly an osseous and partly a fibrous or fibro-cartilaginous consistence, are very difficult to make; but a thin shaving may generally be removed with a strong knife, used in such a manner as to cut through both the fibrous and ossified portion if the latter is not very

hard and abundant. This operation is sometimes rendered easier if the tumour be dried. After a section has been removed it is to be re-moistened with water and may then be soaked and mounted in glycerine. As I have stated, under the examination of hard tissues (page 108) a far better plan is to soak the hard tissue in glycerine for a considerable time, in fact until it loses its brittle character, and may be cut with a strong sharp knife. Sections thus obtained may be examined in glycerine. The calcareous matter may be dissolved out with hydrochloric acid, in the same way as in investigations on bone and teeth, and by partial solution with very dilute acid most instructive specimens may be obtained.

352. Myeloid.—This term was applied by Sir James Paget to a soft pulpy growth which probably has its origin in the bone itself.

It often presents many of the characters of soft cancer, but its mode of growth, its history, and its anatomical appearances distinguish it from tumours of this description. Cells differing considerably in shape and size but containing a vast number of bioplasts are present in myeloid tumours. The bodies in question closely resemble certain cells which were originally described by Professor Kölliker, and are found in considerable number in the medullary cavity of fœtal bones, and to a less extent in the same situation and beneath the periosteum even in adults.

Myeloid tumours are not uncommonly found in connection with the jaw-bones, and certain forms of *epulis* have a myeloid structure. Sir James Paget has described such tumours connected with the bones of the skull, and they are not uncommon in those of the lower extremities. The microscope characters have been very carefully considered by the late Mr. Gray,* and an excellent example of the disease has been reported by Mr. Hulke.†

353. Examination of Striped Muscle.—The best muscular fibres for examination are to be obtained from the thinnest muscles of small animals. The very delicate cutaneous muscles, the pectoral or the sartorius, or the thin abdominal muscles of the frog, hyla, or newt, the diaphragm of the rat or mouse, particularly the white mouse, or the tongue, or the auricle of the heart of any small animal will furnish good specimens of elementary fibres of striped muscle.

A small piece of the fresh tissue may be snipped off with fine sharp scissors, placed upon the glass slide, moistened with serum, vitreous humour, or a one per cent. solution of chloride of sodium, and covered with thin glass, allowed to press only very gently upon the specimen

* "Transactions of the Medico-Chirurgical Society," 1856.

† "On Tumours connected with Bones."--("Archives of Medicine," No. II, page 104, and Plate XIII.) See also a memoir by C. Robin. "Sur l'Existence de deux espèces de Nouvelles d'Eléments Anatomiques qui se trouvent dans le Canal Médullaire des os." Paris, 1849.

which is to be examined first under an inch, next under a quarter, and lastly under a twelfth of an inch object glass.

The general arrangement and form of the fibres in voluntary muscles is well shown in a transverse section of the pectoral muscle of a teal (*Querquedula crecca*), which has been put upon the stretch, and allowed to become perfectly dry. A section cut as thin as possible may be re-moistened with water, and examined in the usual manner. The position of the vessels, their relation to the fibres, and the character of the capillary network are easily demonstrated in specimens which have been injected with transparent Prussian blue or carmine injection.

For displaying the vessels of muscle, injected specimens must be prepared according to the directions given in Chapter VII, and the bioplasts must be stained with carmine if it is desired to study their arrangement. The mode of growth of the tissue and the manner in which new fibres are formed can be studied in specimens so prepared. The transverse striæ are seen most distinctly in muscular fibres which have been for some time preserved in glycerine. If it is desired to study the contraction of the fibres, the mode of proceeding recommended in p. 131, should be adopted.

The ultimate fibrillæ are well displayed in the muscles of many of the cartilaginous fishes, especially the lamprey. The cleavage can be very satisfactorily effected and the "ultimate sarcous particles" separated from one another in the muscular fibres of the eel, young newt, salamander; and those of the cameleon, and the proteus may be also recommended.

354. Sarcolemma.—The muscular fibre of the skate, as Mr. Bowman has shown, is remarkably well adapted for demonstrating the sarcolemma. The sarcous matter may be ruptured while the investing sarcolemma remains entire, and may be thus easily perceived even when it is extremely thin. A few of the long muscular fibres from the fin may be spread out upon a piece of glass with the aid of needles. In this operation it will often be found that the rupture of the sarcous matter in the interior of several fibres has taken place. In the muscles of insects the sarcolemma covered with tracheæ and nerve fibres is also successfully demonstrated. See my paper on this subject in the Transactions of the Microscopical Society for 1864.

355. Branched Muscular Fibres.—Several modifications of striped muscle have been described of late years, and it is desirable to consider the best methods of demonstrating a few of the most important of these. Branched muscular fibres and fibres arranged to form networks exist in abundance in every part of the heart. Fibres of this nature are also found in the tongue of the frog (as was pointed out by Kölliker), from which organ they may be obtained as follows: the tongue is to be separated from the animal, and boiled for a few

moments in water; the mucous membrane is cautiously dissected off from a small portion, and a few minute pieces are to be carefully snipped off with scissors from the edge of the tongue, just beneath the mucous membrane. These are to be torn with very delicate needles, and then examined with a quarter of an inch object-glass. In this manner very perfect fibres may generally be found; but care must be taken not to boil the tongue for too long a time, for the fibres might thus become too brittle to admit of separation. They may be also obtained and more clearly demonstrated in the case of tongues which have been soaked for weeks in glycerine containing five drops of acetic acid to the ounce.

These branched fibres from the tongue are very beautiful. In good specimens they are seen to ramify after the manner of the branches of a tree, gradually becoming thinner, until each terminates in a delicate extremity, which is of a tendinous nature, and is incorporated with the sub-mucous areolar tissue or *corium*. The transverse striæ may be observed in the thinnest branches, but cease some distance from the terminal extremity of the fibre. Branched fibres also exist in the upper lip of the rat.* The gradual tapering of the muscular fibres of the tongue towards their small tendons which are inserted into the corium, has been well described by Dr. Hyde Salter.†

The nerve fibres form networks amongst the delicate ramifications of the muscular fibres. Neither upon the fine branched fibres of the tongue nor upon those of the heart are any terminations or terminal organs of any kind to be demonstrated. Networks of the finest nerve fibres can however invariably be discovered in properly prepared specimens.

Amongst the matters vomited by patients suffering from certain affections of the stomach, beautiful specimens of striped muscular fibre may often be found; and in the evacuations of cholera patients, such specimens were almost constantly observed. In the stomach, the fibres sometimes break up into the discs described by Bowman, and I have obtained these discs produced by transverse cleavage, by macerating the muscles of a fœtus for a long time in strong acetic acid, and also from muscular fibres of reptiles (frog, newt, snake, cameleon, crocodile, tortoise) long kept in glycerine.

Among the reagents of use in investigating the structure of muscular fibre, are a dilute solution of caustic soda, acetic acid, and very dilute solutions of chromic acid or bichromate of potash in glycerine. Preparations of muscular fibre may be preserved moist in glycerine,

* Huxley: "British and Foreign Medico-Chirurgical Review," 1853, No. XXIV, page 313.

† Article "Tongue," Dr. Todd's "Cyclopædia of Anatomy and Physiology."

glycerine jelly, chromic acid, or solution of creosote, or they may be dried and mounted in Canada balsam.

356. Examination of Muscular Fibre in a state of Fatty Degeneration.—This most interesting subject was originally investigated by Vicq. D'Azyr. Lately it has received the attention of many observers, both in England and also on the Continent. The memoir of Dr. Quain, "Medico-Chirurgical Transactions," vol. xxxiii, contains a most excellent account of fatty degeneration of muscular fibre.

The muscular fibres of the heart very commonly undergo this change, and it is frequently well marked in the musculi papillares, to which the tendinous cords of the auriculo-ventricular valve are attached. Almost any muscles which have long been out of use, as happens in many cases of paralysis, and in numerous forms of club-foot, will be found to exhibit it also.

Mr. Gant showed that many of the muscular fibres of the heart of animals which had been fatted for the Cattle Show, had undergone fatty degeneration. "Evil Results of Over-feeding Cattle," by F. G. Gant (Churchill, 1858).

A beautiful specimen of muscular fibre in a state of fatty degeneration is represented in fig. 8, pl. LIX, from one of the abdominal muscles of the Hyla. The contractile tissue is seen to be nearly entirely replaced by large oil globules.

Dr. Meryon mentions the case of four brothers in whom all the muscles of the body lost their contractile power. In this family the females were unaffected. "Medico-Chirurgical Transactions," vol. xxxv, p. 73.

Sometimes there is not the slightest appearance of transverse striæ on the fibre, which appears to be composed of rows of minute and highly refracting globules of oil. Sir James Paget noticed that the nuclei of muscular fibres in a state of fatty degeneration, were very often absent. In other specimens, no distinct globules can be seen, but the whole fibre appears made up of granular matter. Fig. 7, pl. LIX, represents two elementary fibres in a state of fatty degeneration from one of the papillary muscles of the mitral valve. It must, however, be borne in mind, that fibres in a state of commencing decomposition exhibit, in a slight degree, this granular appearance.

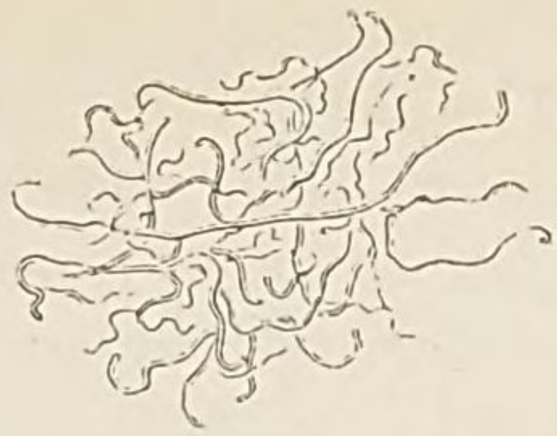
Fibrous Degeneration.—Not unfrequently fibres of muscles undergo gradual conversion into fibrous tissue, a change which is also particularly common in the small papillary muscles of the heart. By carefully tearing up a fragment with needles, the fibrous structure can be clearly made out.

The examination is conducted in a similar manner to that of healthy muscle; very small pieces may be cut off with scissors, and the elementary fibres carefully separated from one another with very fine needles.

Fig. 1.

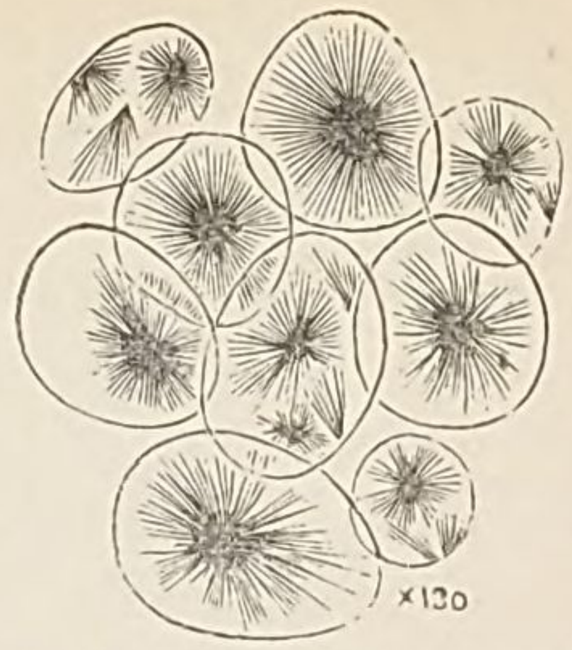
Hypertrophied areolar tissue, showing coarse white fibrous tissue in great quantity. × 215. p. 374.

Fig. 2.



Fibres of yellow elastic tissue, from the scrotum of a man operated on by Sir W Fergusson. In this case the areolar tissue had undergone considerable hypertrophy × 215. p. 374.

Fig. 3.

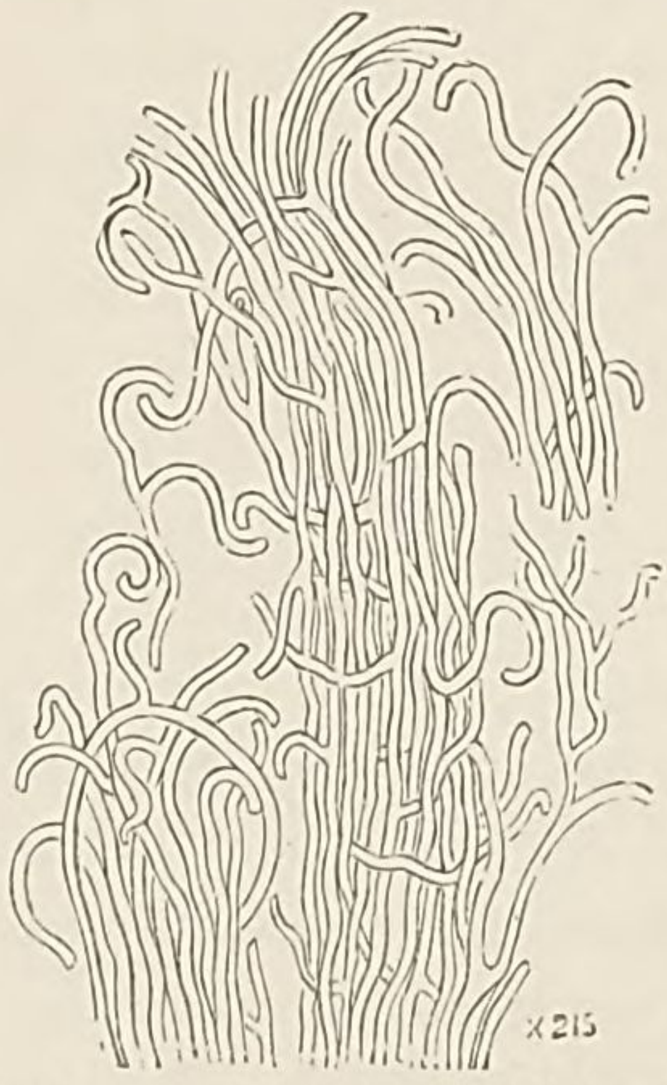


Large fat vesicles, with crystals of margarine, from a large tumour connected with the testicle, removed by Sir W. Fergusson. × 130. p. 375.

Fig. 4.

White fibrous tissue, unravelled from a tendon. p. 374.

Fig. 5.



Yellow fibrous tissue, the fibres of which have been unravelled; from the ligamentum nuchæ of a sheep. p. 374.

Fig. 6.

Adipose tissue, with vessels injected, from the capsule of the kidney of a dog. × 130. p 375

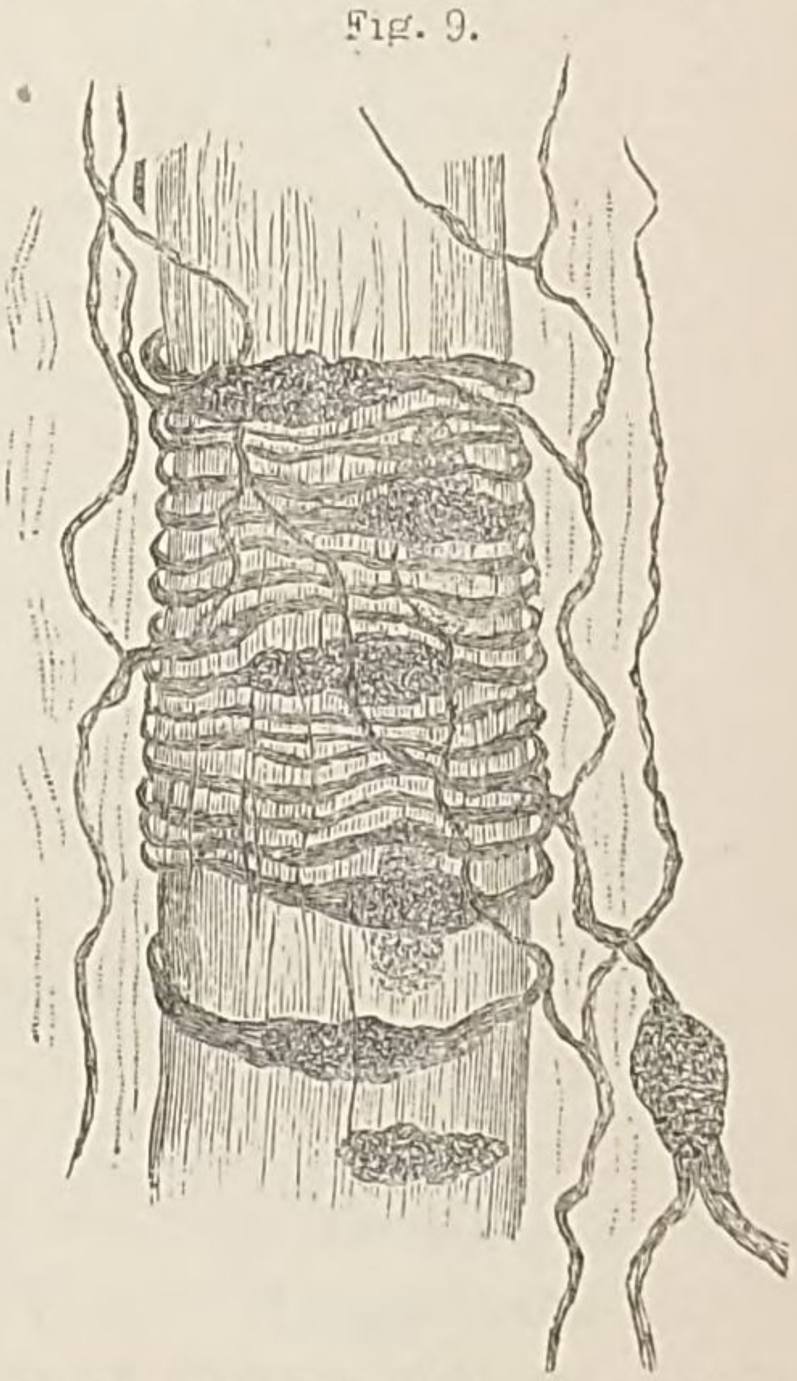
Fig. 7.

Muscular fibre, fatty degeneration, from the heart × 215. p. 380.

Fig. 8.

Muscular fibres in a state of extreme fatty degeneration from prolonged inaction. Also capillaries and nerve fibres. From a hyla which had been long in confinement. × 220. p. 380.

Fig. 9.



Small artery. Hyla. Showing muscular fibres and nerve fibres. × 700. p. 381.

Fig. 10.

Muscular fibres, unstriped, from the human small intestine. × 215. p. 381.

$\frac{1}{1000}$th of an Inch |___| × 215.

" " |_____| × 700.

[To face page 380

The addition of acetic acid causes the oil globules to become more distinct. The oily nature, both of the globules and granules, may be proved by the addition of a drop of ether, which dissolves them. Upon the evaporation of this ethereal solution, a globule of oil remains behind, which will be found to leave a greasy stain when rubbed upon a piece of clean glass. This reagent also enables us to distinguish between globules of oil, and globules of phosphate of lime, which often much resemble each other in form, colour, and refractive properties.

357. Examination of Unstriped Muscle.—Involuntary, smooth, or non-striated muscular fibre may be obtained from various situations, both in man and also in the lower animals. These fibres are most abundant in the alimentary canal, the uterus, the bladder, the ducts of glands generally, the arteries and veins, particularly the smaller branches, and they are also found dispersed amongst fibrous tissue in certain situations, particularly in the skin. The bundles of pale muscles connected with the hair bulbs may be very readily demonstrated in some of the smaller animals; connected with the large hairs about the mouth they are very distinct. The elongated cells, of which this form of muscle is composed, are also to be demonstrated in the trabecular tissue of the spleen, and corpora cavernosa penis, the urethra, &c. Involuntary muscle, which has hitherto been described as consisting of flattened bands, was proved many years ago by Kölliker, to consist of the elongated bodies just referred to. The contractile fibre cells have been arranged in three classes:—1. Short, rounded, or flattened cells, somewhat resembling epithelium. 2. Flattened bands, with fringed edges. 3. Long rounded or fusiform fibres, slightly wavy, and terminating at each end in a point. To these must be added, 4. Fibres with three or more contractile threads radiating from a common centre.

The first two varieties are obtained from the blood-vessels. The third form is met with in the intestinal canal, uterus, &c., and the last in the bladder of the frog, newt, and other animals. The fibres may be readily isolated by macerating small pieces of the muscular coat of the alimentary canal, &c., in dilute nitric acid, containing about twenty per cent. of strong acid. By a little teasing, with the aid of fine needles, separate cells may be readily obtained. Fig. 10, pl. LIX, represents some of the contractile fibre cells from the ileum. These cells may also be demonstrated without difficulty in the intestine of any small animal—that of the white mouse being most favourable, and the uterus of the same animal especially when quite young will furnish beautiful specimens.

The small arteries of the frog or newt may be distended with transparent injection so as to put the muscular fibres upon the stretch. Here and there rupture will occur and in these situations the cells will be more or less separated from one another, and individual muscular fibre-cells

may by this method be isolated, fig. 9, pl. LIX. But of all the situations in which to study the arrangement of the fibres of organic muscle, the most satisfactory is the bladder of the frog. The vessels should be injected with Prussian blue fluid and the bioplasm stained according to the method described in p. 104. Not only can each individual muscular fibre cell be traced from one end to the other, but the distribution of the nerve fibres can be distinctly followed. In the tissue indicated many muscular fibre cells having a triangular central part with three fibres radiating from it will be found.

Fatty degeneration of the fibre cells of organic muscle may be studied in the uterus after delivery, or in small arteries in many cases of disease in which vascular degeneration has taken place, as for example, in chronic renal disease, or in those cases of general fatty degeneration which result from many years of injudicious over eating, or alcohol drinking, or both.

358. Examination of Nerves.—The general anatomy of the trunk of a nerve is demonstrated without difficulty. It is better to take a fine fibre and tear it up with very fine needles upon a glass slide. After the addition of a drop of serum, it may be covered with thin glass. The delicate nerve fibres of any small animal, or the ciliary nerves of the eye furnish good specimens. The nerves of the frog are very large, and in them all the essential structures of nerve fibres may be very distinctly demonstrated. Glycerine is a good medium for the examination of nerve fibres. Observations should, however, be made upon specimens prepared according to different methods. In this way only can the student hope to avoid drawing erroneous inferences.

If a nerve be placed in a little water, a curious change takes place. The constituents of which the medullary sheath (white substance of Schwann) is composed, become altered so as to exhibit two distinct lines,. or a *double contour*. This change probably depends upon the fatty matter of the myelin being partly separated from the albuminous material with which it was incorporated. Although the double contour line is undoubtedly produced by soaking in water, the existence of a special highly refracting material within the tubular *membrane* and around the *axis cylinder* in large fibres from the spinal nerves cannot be questioned. It is instructive to compare the appearance of the white substance in water with that produced in highly refracting media as syrup or glycerine.

If dark-bordered nerve fibres near their point of distribution be examined after having been for some time soaked in glycerine and acetic acid (five drops to the ounce) fine nerve fibres which are quite invisible in specimens immersed in water or serum, may be demonstrated with the greatest distinctness. In not a few instances, one of which I have figured in a paper in the Phil. Trans., for 1862, pl. XLII,

fig. 11, a single dark-bordered fibre may be seen gradually tapering towards its ultimate distribution. Running by its side, in the same sheath, as some would say, is one of these fine fibres, which, after all, are but the continuation of dark-bordered fibres much nearer to their ultimate distribution than at the point where the dark-bordered character exists. A dark-bordered fibre often divides, and near their distribution each of its subdivisions divides into a finer dark-bordered fibre and a pale fibre, or into two pale fibres, fig. 1, pl. LX.

The general distribution of the nerves may be well seen in the ear of the mouse, from which the thin skin covering it has been carefully dissected off. In the dura mater also, even in man, I have seen many individual nerve fibres arranged so as to form with others a coarse network, and a single fibre may often be traced for a very long distance.

Beneath the skin of the eyelid of the frog, a wonderful plexus of dark-bordered nerve fibres will be found, the study of which is not difficult especially in specimens which have been soaked in glycerine and acetic acid (five drops to the ounce).

Fibres in the above tissues may be seen to leave their companions and pass a short distance in company with others, so that an intricate network is in this manner formed. The mesentery of the mouse is a very favourable structure in which to trace the finest subdivisions of dark-bordered nerve fibres.

For investigating the ultimate networks of very fine nerve fibres there is no tissue like the bladder of the frog, which is so very thin that all the anatomical elements entering into its formation can be studied *in situ*, fig. 1, pl. LX. (See my paper in the Phil. Trans., vol. 152, p. 889, 1862.)

The papilla of the tongue of the frog and especially of the hyla, are very favourable for studying the arrangement of the ultimate nerve fibres upon a sensitive surface. (See my memoir on the Frog's tongue, &c., Phil. Trans., 1864, vol. 155, p. 443.) Here a network of excessively fine fibres may be demonstrated with great distinctness. In the olfactory mucous membrane, in the retina, in the Pacinian corpuscles the question of the mode of termination of nerve fibres and their ultimate ramifications may be investigated with advantage.

In all cases, bioplasm (nuclei) are found in connection with the terminal ramifications of nerve fibres. From this oval or spherical mass of living matter, the fibre grows, and the current which traverses the fibre is probably set free upon its surface, whence it is transmitted by the fibres which grow from the bioplasm. Terminal extremities have been described both at the peripheral distribution of nerves and in nerve centres. "Ends," it is affirmed with the greatest confidence, constitute the natural arrangement as regards all nerve instruments. I have never succeeded in demonstrating an end or a terminal organ of any kind in connection with any nerve apparatus, and have advanced numerous

arguments grounded upon facts demonstrated by myself and shown to others during the past twenty years, in favour of the view that nerve fibres with the "cells" with which they are connected form complete circuits. See "Bioplasm," an introduction to Physiology and Medicine. The ultimate nerve fibres, × 1700, just beneath the epithelium of the mucous membrane of the human epiglottis are well shown in fig. 2, pl. LX.

The minute structure of nerves becomes much altered by division in the living body, by stretching or pressure. In fact all trace of structure may be lost, the bundle of nerve fibres assuming the appearance of fibrous tissue with lines of oil globules which have resulted from changes in the white substance of the medullary nerve fibres. A beautiful instance of fatty degeneration of nerve fibres, precisely corresponding to the change which takes place in fatty degeneration of the muscular fibres of the heart, occurred some years ago in a case of Dr. Todd's, in which the pneumogastric nerve had been much stretched and had, indeed, become gradually incorporated with the walls of a large aortic aneurism. In this instance each individual dark-bordered nerve fibre could be made out by the oil globules and granules that had been left in the tubular membrane of each fibre.

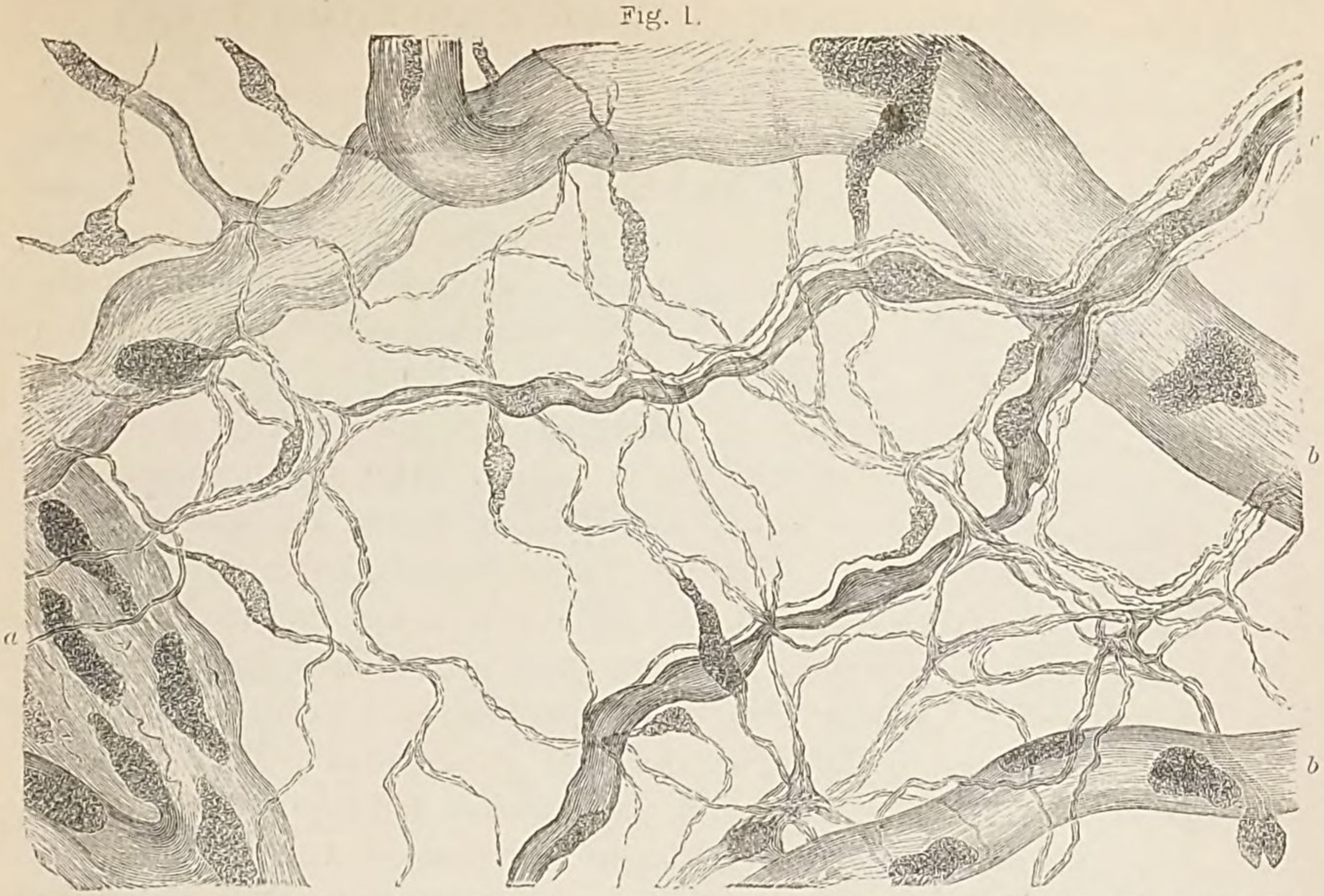
Microscopical Examination of Organs.

359. Heart.—The muscular fibres of the heart exhibit the transverse striæ characteristic of striped muscle. The fibres are arranged in long bands, and upon carefully examining a well-prepared specimen, taken either from the heart of man or an animal, frequent and distinct anastomoses and branchings of the fibres—in short, a net-work will be observed. The arrangement is beautifully distinct in the thinnest part of the auricle of the heart of the frog, and especially of the hyla. In the latter also the finest nerve fibres, arranged to form delicate net-works upon and amongst the muscular fibres can be very clearly demonstrated.

In order to exhibit the general course of the fibres of the heart, that of man or of the calf or sheep should be boiled in water until it is quite firm. It may then be preserved in alcohol. Layers of fibres may then be detached and these may be further torn up and separated into finer fibres and laminæ by the aid of needles.

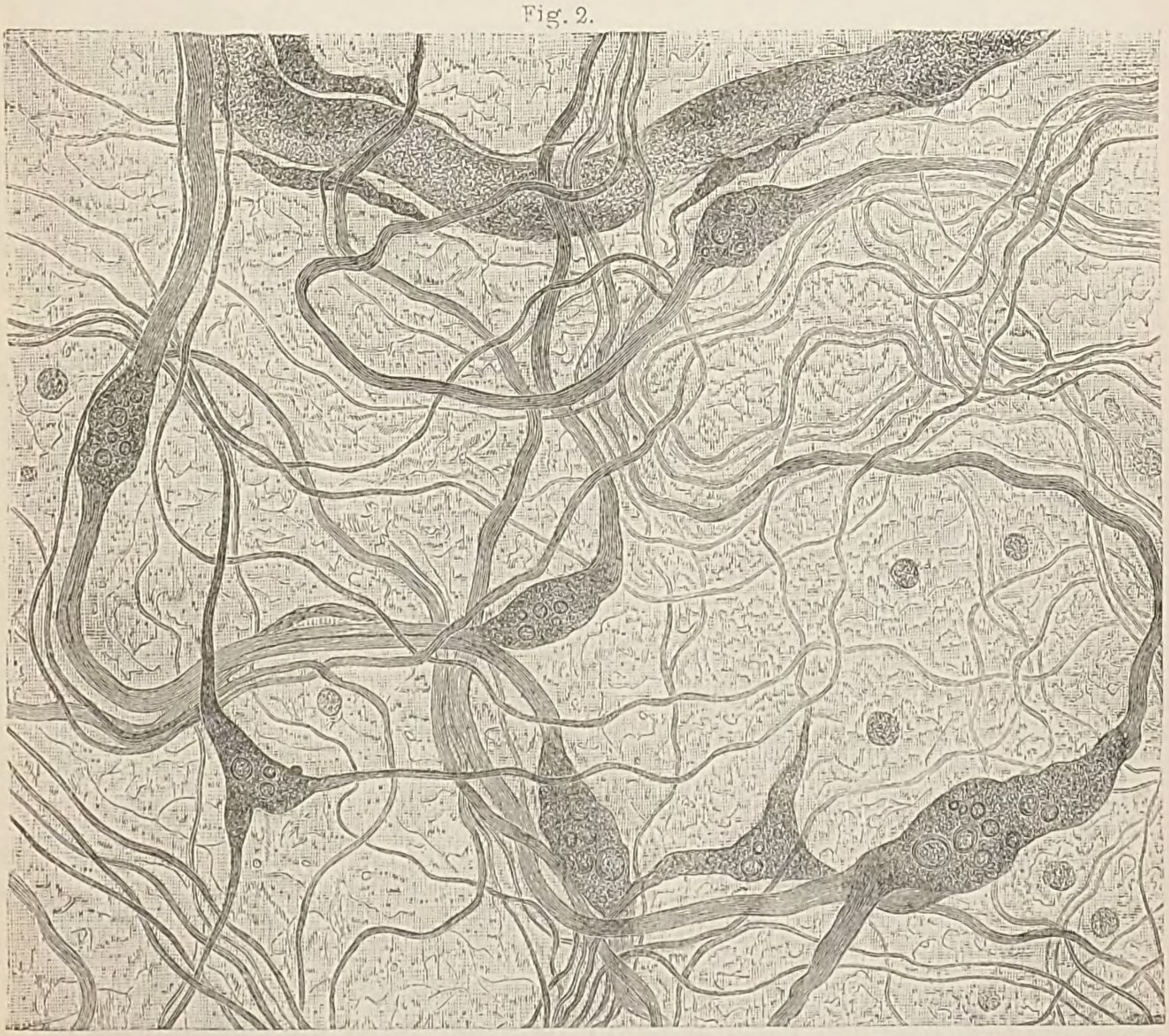
But the method which has been most successful in my hands is that which I have fully described in Chapter VII, p. 103. In specimens thus prepared, the observer will, by making transverse sections, be able to demonstrate the important fact that the bioplasm of the muscular fibres of the heart is like that of many of the muscles of insects and all (?) the muscles of embryos (human and others), in the centre of the fibre. The new tissue is found immediately around the bioplast in the

Fig. 1.



Nerve fibres distributed to the bladder of the frog. *a*, vein with numerous bioplasts in the walls. *b*, *b*, capillaries. *c*, dark-bordered nerve fibre, dividing into two branches, the upper one of which is seen to pass into pale fibres which form a network running in every part of the specimen Fine fibres like the last run with the dark-bordered nerve fibre. × 700 p. 383.

Fig. 2.



Mucous membrane of the human epiglottis just beneath the epithelium A small portion of Fig. 1, pl LXII (× 1700). The bioplasts connected with the finest nerve fibres and the manner in which the latter ramify are well seen. No true terminations or "ends" are to be found. × 700. pp. 384, 396. See also fig. 1, pl. LXII., p. 398

$\frac{1}{1000}$th of an Inch ⌊——⌋ × 700.

" " ⌊————————⌋ × 1700. [To face page 384.

central part of the fibre, while the oldest portion of the tissue is at the circumference where disintegration is taking place.

The muscular fibres of the heart, like other forms of striped muscle, are liable to the two forms of degeneration already referred to—the *fatty* and the *fibrous*. The first condition is not uncommonly found in those who have long suffered from chronic bronchitis, gout, emphysema, and some other conditions. Several muscular fibres of the musculi papillares and of the ventricles near their insertion into the chordæ tendineæ may undergo this change without those of the heart generally being involved—but cases occasionally are met with in which not a single healthy muscular fibre can be found in any part of the heart.

360. Arteries and Veins.—The examination of the coats of the larger arteries and veins must be conducted upon thin sections cut longitudinally and transversely. Very instructive preparations may be obtained from the aorta in cases of commencing atheroma. If small pieces be prepared as directed in p. 103, numerous small bioplasts will be seen growing and multiplying amongst the muscular fibre cells and elastic fibres of the vessel. It is by the growth of these bioplasts that the laminæ are separated from one another and the coats of the vessel rendered weak and brittle. After such changes have continued for a considerable time, and they often go on for several years, many of the bioplasts die and become disintegrated. The granular matter left consists of fatty matters, earthy salts, and albuminous substances which constitute "atheromatous" or cheesy matter.

The atheromatous deposit contains numerous oil globules, with granular matter, partly fatty and partly albuminous, aggregations of minute fat globules, and much cholesterine, pl. XXI, fig. 1, p. 190, which has gradually crystallized from its solution in the oily fatty matter. Cholesterine is an invariable constituent of the fatty matter found in all organs in a state of true fatty degeneration. Its presence may be detected by evaporating an alcoholic solution of the fatty matter present in the tissue.

The cholesterine which has slowly crystallized from the fat in the coats of an artery during life may exert an influence upon the further deposition of atheromatous material, and in this way we may account for the large accumulation often met with.

Besides the atheromatous deposit, plates of calcareous matter, resembling bone to the unaided eye, are often found in the coats of arteries and occasionally these form a complete ring round the vessel. The calcareous plates contain neither lacunæ, nor canaliculi, nor it need scarcely be said Haversian canals.

The coats of the larger veins are to be examined according to the same plan as has been recommended for the arteries.

361. Examination of the Smaller Vessels.—The smaller vessels

may be examined entire, and present beautiful objects for microscopical observation. A portion of the mesentery of a child, or of one of the lower animals, or a small piece of the pia mater may be selected; or one of the smaller arteries of the brain may be freed from cerebral matter by gentle washing in glycerine and water, and placed in the microscope, with the usual precautions. If the specimen be treated with a drop of acetic acid, the nuclei of the contractile fibre cells, and, in some cases, also those of the epithelial cells, on the lining membrane are brought into view. The student will observe a remarkable difference in the diameter of the small arteries at different points. The actual changes may be studied in the arteries of the foot of the living frog as described in p. 133. But in many specimens prepared by injecting the carmine fluid very soon after death, and then the Prussian blue fluid, according to the directions given in p. 104, these alterations in the diameter of the arteries may be produced and preserved permanently. The pia mater of the lamb or sheep is favourable for the investigation. Arteries will be found whose diameter differs remarkably in a length of a few hundredths of an inch. The thickness of the coats varies, of course, in like manner, so that in one place the walls of the vessel may be three times the width of its cavity, while a very short distance above or below this point the internal diameter may be six times as great as that of the wall. Here the artery appears to be all firm, thickened tissue; there it appears as a tube, with very thin membranous walls. The investigation of such changes in healthy arteries is important with reference to the discussion concerning the nature of the thickening observed in arteries in certain cases of long-standing disease, figs. 4, 5, 7, pl. LXI.

Small Vessels of the Kidney, Brain, and other Organs.—A thin section of the cortical substance of the kidney often displays the minute vessels well. The examination of the vessels in this organ is of especial interest, because they are known to undergo important alterations which could not be detected unless the observer were previously well acquainted with their appearance in health. If a section of a healthy kidney is to be examined, with a view of observing the characters of the minute vessels, it will be better to wash the preparation previously in glycerine and water, with the aid of the wash-bottle, fig. 6, pl. XVI, p. 168, in order to remove much of the epithelium of the renal tubes. Upon the addition of a drop of acetic acid the vessels are at once brought into view. It will now be noticed that the coats of the veins everywhere appear to be very thin, being represented only by a defined line on each side of the vessel, while the arteries are at once recognized by the greater thickness of their walls, and by the distinct arrangement of the bioplasts of the circular and longitudinal fibres. All these points may, however, be observed much more satisfactorily in sections re-

moved from a specimen which has been injected with carmine fluid, and subsequently with Prussian blue fluid (page 105).

The coats of a healthy Malpighian artery appear to be about the fifth or sixth part of the total diameter of the vessel, but in disease the vascular wall may have increased in thickness to such an extent as even to equal in width the canal itself, and to be as much as one-third of the total diameter of the vessel, fig. 2, pl. LXI. The most perfect specimens of this morbid condition are to be obtained from the small contracted kidneys of intemperate persons. Much of the thickened tissue consists probably of the altered muscular fibre cells which have degenerated into a form of fibrous tissue. Fig. 7, pl. LXI, represents the appearance of a portion of a Malpighian artery considerably thickened, but not to such a degree as often occurs. The thickening of the walls is well shown in injected specimens.

Minute Vessels of the Brain.—A knowledge of the appearance of the minute arteries of the brain in a state of health is especially important to the morbid anatomist, because in disease these vessels are not unfrequently found to have undergone very important changes, which must necessarily much interfere with the discharge of their functions. In cases of white softening of the brain, the small arteries and capillaries are much altered. If we tear away some of the smaller vessels from the softened portion, and, after washing them in glycerine and water, place them in the microscope, we shall often find at short intervals, collections of minute oil globules, easily distinguished by their high refracting power, pl. LXI, fig. 1. These may form small aggregations of globules at short intervals, or may extend entirely round the vessel for some distance. With regard to the exact relation of the bioplasts to the wall of the capillary, there is room for some difference of opinion. If they are not situated upon the internal surface of the wall of the vessels, they certainly project into the cavity, in such a way that the blood corpuscles must impinge against them one after another, as they are driven along the capillary vessels. Some observers consider them to be in its substance, while others contend that they are always external to the vessel. Frequently small collections of oil globules are found on opposite sides of the tubes, alternating with each other, and occupying the position of the bioplasts concerned in the development of the vessels. This condition has been termed fatty degeneration of the vessels, and is probably very similar to the corresponding change in muscular fibre. Fig. 3 represents the characters of a small artery with the commencement of the capillaries from a healthy brain, magnified in the same degree as the companion morbid specimen represented in fig. 1. In many cases of white softening, the small arteries of the brain are entirely surrounded by spherical aggregations of small oil globules which sometimes extend around the vessel to a distance equal to twice

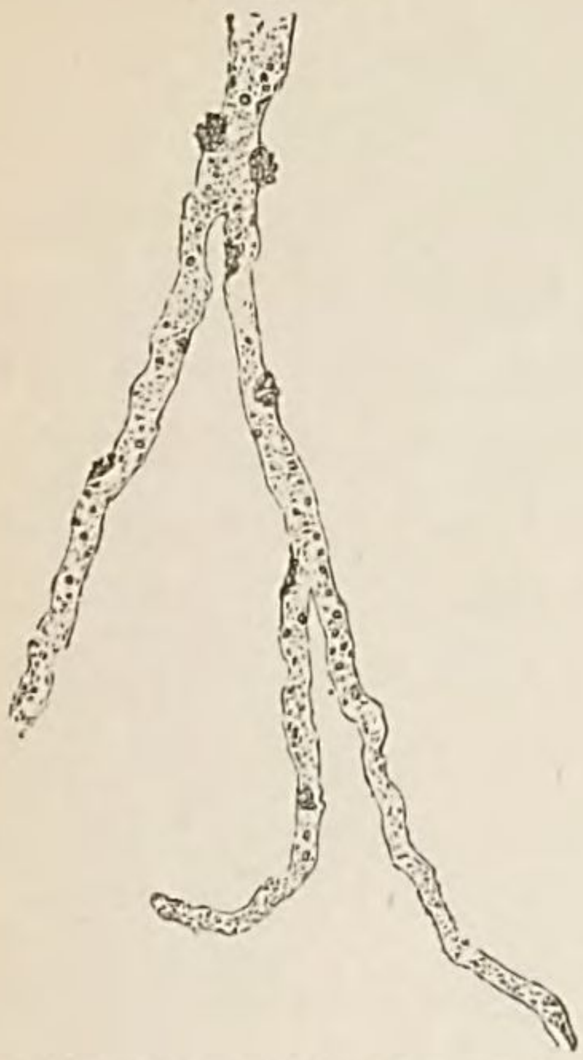
its entire diameter. In such a case it is clearly impossible that the cerebral tissue can be properly nourished. In some instances the passage of the blood through a vessel is prevented by its tube being plugged up with a portion of fibrinous matter. In consequence of this obstruction, the nerve tissue suffers in nutrition, the vessels degenerate, and other changes result.

Morbid changes are found to pervade the capillary vessels of many organs of the body. Thus, the vessels of the retina and choroid are frequently affected so that their walls become softened, and undergo small dilatations at varying intervals, in which the blood seems to collect; and after a time to become stagnant. The little clots increase by the deposition of more clot until the vessel is completely obstructed. The changes which occur in the capillaries of the papillæ of the skin, of the villi, of the liver, kidney, and other glandular organs, are well worthy of attentive study.

362. Nerve Fibres to Small Arteries, Veins, and Capillaries.—The student must not omit to consider the wonderful changes in the distribution of the blood, occasioned through intervention of the nerve fibres distributed to the small arteries, capillaries, and veins. The nerve fibres to the capillaries, first described by myself, in the frog, newt, hyla, bat, and in other animals, are probably afferent or excitor in their action, while those distributed to the muscular fibre cells of the coats of the small arteries and veins exercise a motor influence, regulating the degree of contraction of the muscular fibre cells, and so determining the calibre of the vessels and the quantity of blood that shall flow through the capillaries in a given time, as well as the rate of its flow. The arrangement of these nerve fibres I have described in my work on "Bioplasm," pages 301 to 322.

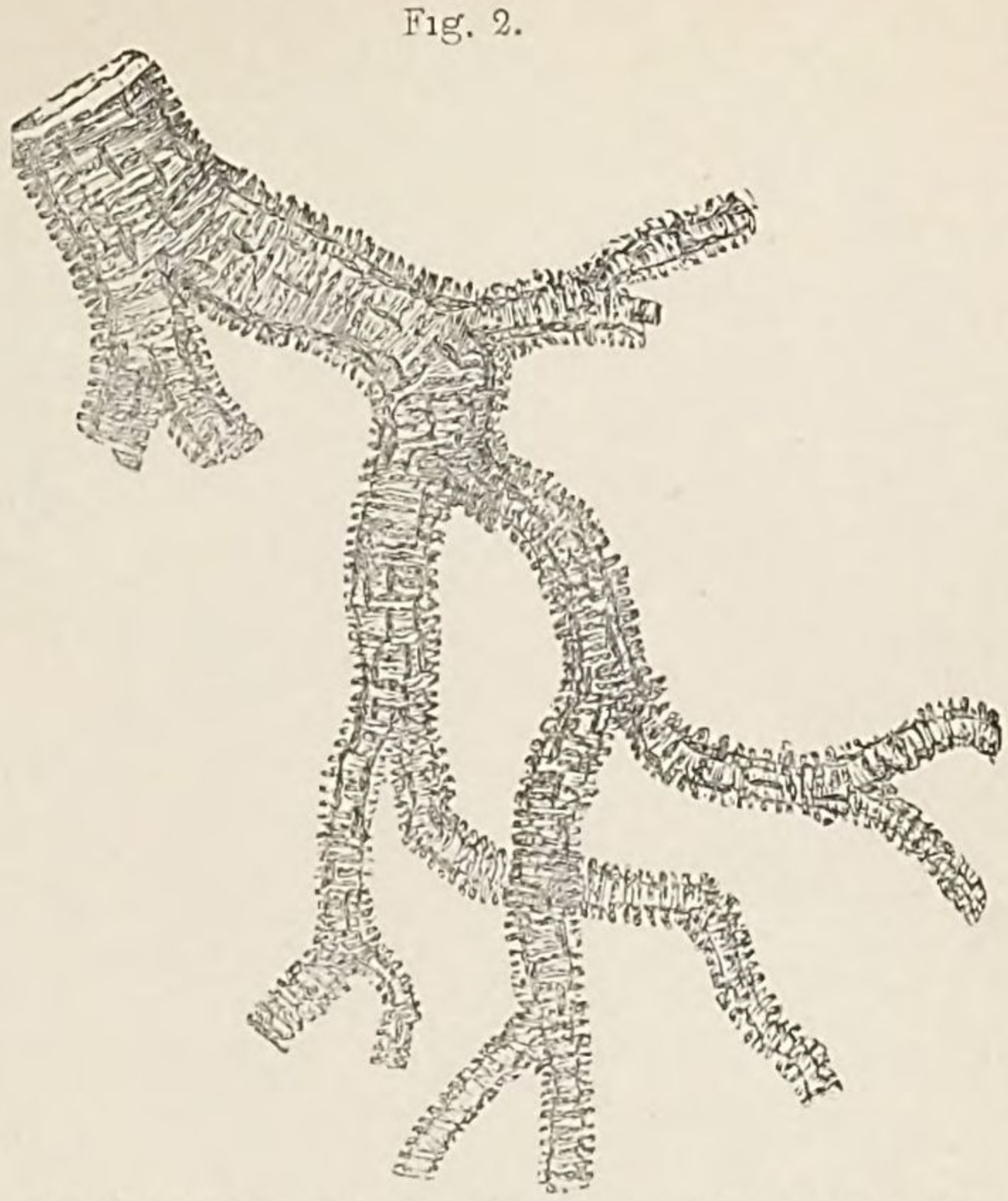
The nerve fibres to the small arteries and veins and to the capillaries are demonstrated very readily in many tissues of the hyla prepared according to the plan recommended in Chapter VII. In the bladder and in the fibrous membranes in the abdominal cavity, in the mesentery, and in such thin muscles as the pectoral, the finest ramifications of the nerves upon the minute arteries and veins and those distributed to the capillaries, may be demonstrated with great clearness. In other situations they can also be discerned. I have made some beautiful preparations of the capillaries of the bat's wing with their attendant nerve fibres, and have thus demonstrated the existence of these in connection with the capillaries of a mammalian animal. I have also distinctly seen nerve fibres upon each side of some of the capillary vessels of the mouse and the mole. In order to demonstrate them, the tissue must be well prepared and exceedingly thin. After the prolonged action of very weak acetic acid the nerve fibres become slightly granular. Until this change has taken place the finest ramifications of the nerve fibres are

Fig. 1.



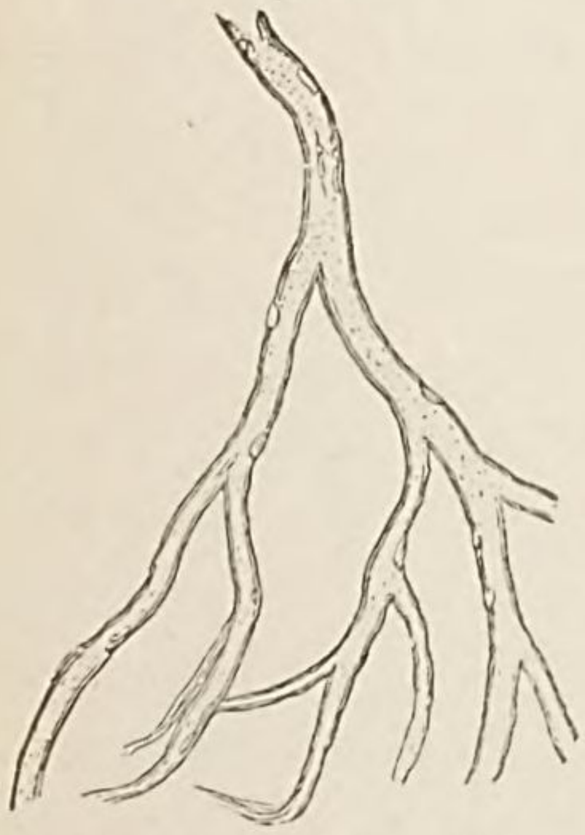
Part of a small artery and capillary vessels in a state of fatty degeneration. Brain. × 215. p. 387.

Fig. 2.



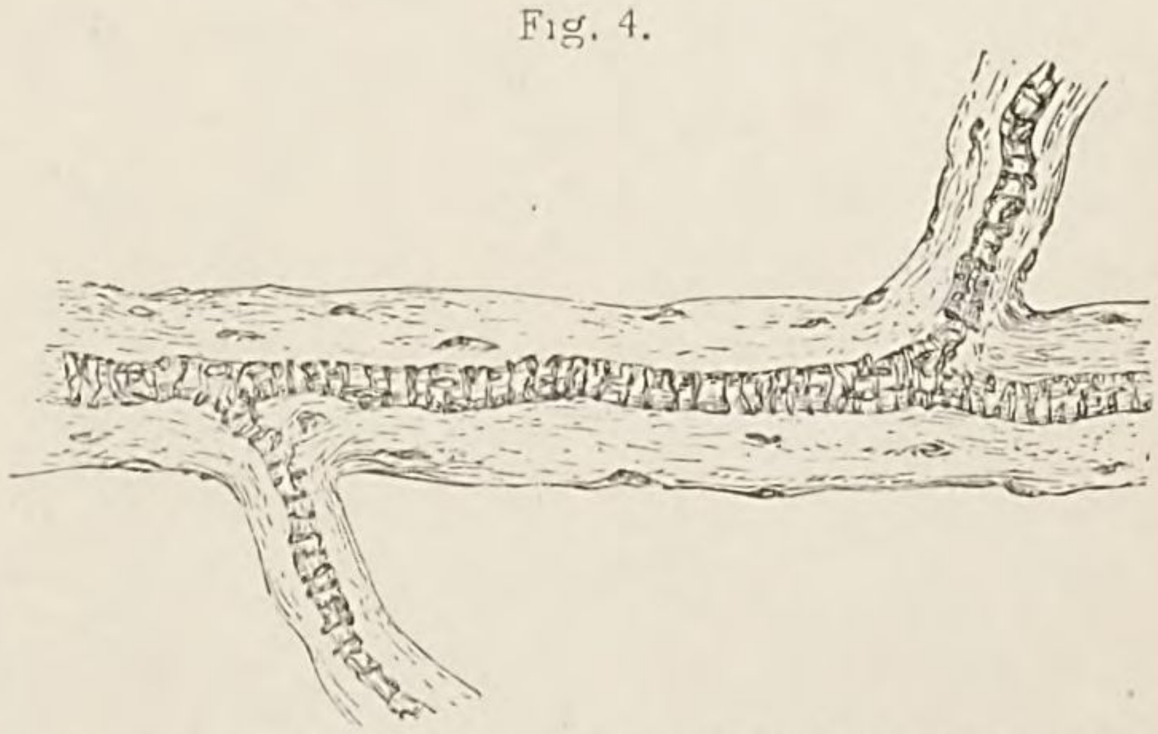
A healthy artery from the kidney of a child 3 years old, showing muscular fibre cells. × 215. p. 387.

Fig. 3.



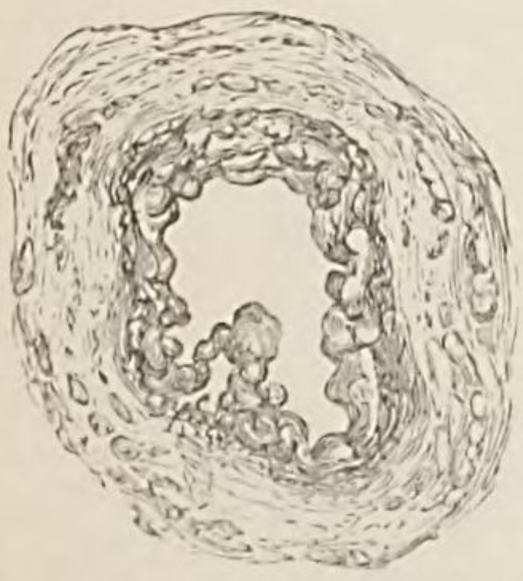
Small artery and capillary vessels from the healthy brain. × 215. p. 387.

Fig. 4.



Artery from the peritoneum of a frog which had been kept without food for some time, showing wasting of muscular fibre cells. × 215. p. 386.

Fig. 5.



Transverse section of an artery from a fatty and contracting kidney, showing complete degeneration of muscular fibre cells, and roughness of internal surface. × 215. p 386.

Fig. 6.

Capillary vessels and nerve fibres, from the skin of the frog. × 215 p 389.

Fig. 7.

Small artery from a kidney. Chronic nephritis. Showing increased thickness of the coats. × 215. p. 387.

$\frac{1}{1000}$th of an Inch ⌊___⌋ × 215.

[To face page 388

quite indistinguishable from the tissues in which they ramify and this is the reason why they have not been before described, fig. 6, pl. LXI.

363. Lung.—To demonstrate the arrangement of the tissues of the lung, small pieces may be cut off and spread upon the glass slide with the aid of needles in the usual way, the preparation being moistened with glycerine and water, or serum. The addition of a little acetic acid causes the yellow elastic tissue to become very distinct. The boundaries and arrangement of the air-cells may also be readily shown.

No opinion with reference to the nature of the walls of the air-cells can be arrived at, unless injected as well as uninjected specimens are examined. The twisted and shrunken capillaries of the recent lung containing a few blood corpuscles, produce an appearance which is very likely to give rise to erroneous inferences with regard to the disposition and coverings of these vessels. The Prussian blue fluid may be employed, and when injection is complete small pieces may be cut off and placed in the carmine staining fluid for four or five hours, and then transferred to glycerine acidulated with acetic acid (ten drops to the ounce).

A most instructive preparation of pulmonary tissue is made by injecting the vessels with tolerably thick transparent gelatine, or with the glycerine and gelatine (page 53), which transudes through their walls, and fills the air-cells. After the lung has been thoroughly injected with the gelatine, it is set aside to get cool. Very thin slices may be made. The vessels will in such specimens be seen *in situ* apparently bare, and uncovered by epithelium.*

Much difference of opinion has been expressed with reference to the question of the existence of epithelium in the air-cells of the lung. I have carefully examined healthy human lungs which have been prepared in various ways, and have completely failed to demonstrate the presence of an epithelial layer as is often figured in books, in the healthy adult, or in the lungs of several mammalian animals. I cannot confirm the representations given in many drawings, in which this epithelium is shown so very distinctly, that one would infer that it was to be seen without the slightest difficulty. In the fœtus and young child, however, particles are found in the air-cells, but it seems to me very doubtful whether these should be regarded in the light of an epithelial covering of the air-cells. The whole subject requires very careful investigation.

364. Trachea and Bronchial Tubes.—The *mucous membrane* of the trachea and bronchial tubes must be examined in the recent state by cutting thin sections with a very sharp knife.

Beneath this mucous membrane is an abundant plexus of lymphatic vessels. In many cases these contain lymph corpuscles and fatty matter

* "Physiological Anatomy," Todd and Bowman, page 393. Mr. Rainey in the "Medico-Chirurgical Transactions," vol. xxxii, 1849, page 47.

in a granular state, so that their arrangement may be easily made out. The lymphatics upon the surface of the lung, immediately beneath the pleura, may also sometimes be very clearly demonstrated. I have one specimen taken from a child, in which these lymphatics are completely distended with large oil globules and granular matter, so that the position of their valves is rendered very distinct, and the smallest branches can be followed into the intervals between the lobules of the lung. In this specimen the tubes certainly form a network, but in many situations appearances are observed which lead to the conclusion that these tubes also commence in cæcal extremities.

In examining the ciliated epithelium of the air-passages, it is only necessary to scrape the surface gently, and, if necessary, the preparation may be moistened with a little serum, as water would very soon stop the motion.

A tough false membrane is formed in many cases of inflammation of the mucous membrane of the larynx and trachea. Occasionally the membranous material reaches the tenth of an inch in thickness, and extends from the *rima glottidis* to the subdivisions of the bronchi of the third or fourth series. Of this I saw a remarkable instance which occurred in the practice of Dr. Cotton of the Seven Sisters' Road. The patient had been taken ill at the time of her confinement, and three or four days afterwards rejected a complete and firm tubular cast of false membrane nearly nine inches in length and quite complete. The material of which it was composed consisted of fibrin with numerous altered and growing colourless blood-corpuscles, and a few epithelial cells upon its deep surface.

Tracheæ, characteristic of the class of insects, may be readily obtained by removing the viscera of a common fly or other small insect, and tearing carefully with needles. Some are figured in pl. XLIII, fig. 3, page 296. They may be examined after the addition of a drop of water, or they may be dried or moistened with a drop of turpentine, or mounted in Canada balsam. The branchiæ of many mollusca (oyster, mussel) exhibit ciliary motion very beautifully. The gills of fish, the lungs of frogs, newts, and serpents, will also furnish many very instructive specimens.

365. Examination of the Lung in a Morbid State.—The same general method of procedure is employed here. The lungs of emphysematous patients are particularly worthy of study. According to the observations of Mr. Rainey, the pulmonary membrane is found perforated with many minute holes, and here and there numerous oil globules may be detected; the fibres of yellow elastic tissue are stretched, and have lost their elasticity; the vessels are much elongated, and the interspaces between them much enlarged.*

* "Medico-Chirurgical Transactions," vol. xxxi, 1848, page 297.

Of *tuberculous lung*, different parts should be submitted to examination. If we find a cavity, its contents, the surface of the walls, and a section of the immediately adjacent tissue should be examined. If the tubercles have not broken down, small portions may be removed upon the point of a knife, moistened with a drop of serum, and examined in the microscope. A tubercular lung which has been injected, affords very instructive specimens. The beautiful preparations of Professor Quekett, showed that the capillaries into which the injection had passed were those of the original air-cells of the lung, and not new vessels belonging to the tubercle.

In pl. XL, figs. 1 to 5, page 280, the general appearance of fragments of elastic tissue of the lung, which are met with in the sputum in many cases of phthisis is given. The characters of different specimens of sputum are considered in Chapter XIV, p. 278.

Crystals of cholesterine are occasionally met with amongst the cheesy matter which makes up the greater part of some tuberculous masses found in the lung after death, especially when these are circumscribed and thus prevented from escaping into the bronchial tubes. If no crystals can be detected, a portion of the mass, placed in a watch-glass, may be treated with a few drops of alcohol. As the alcohol gradually evaporates, beautiful crystals of cholesterine form, and may be subjected to microscopical examination in the usual manner.

The small, white, calcareous masses, which are not unfrequently met with in the lungs of phthisical patients, pl. XXXIX, fig. 3, page 276, and which are from time to time expectorated in sputum, may be examined as opaque objects, with low powers; or fragments may be broken off, and subjected to examination. After having been dried, they may be placed in turpentine or Canada balsam. If we test the particles with a drop of acetic acid, we shall find that they dissolve with effervescence, showing the presence of carbonate. One part of the acetic acid solution may then be treated with excess of ammonia, when a precipitate of phosphate of lime will be immediately thrown down. The presence of lime may be detected in the other portion of the acid solution, by adding to it a little solution of oxalate of ammonia.

Lung in Pneumonia.—If the examination may be conducted at once while the tissue is fresh thin sections may be placed in serum or vitreous humour, or in a 2 per cent. solution of chloride of sodium. With care small portions of pneumonic lung may be stained in the carmine fluid (p. 104), and afterwards preserved in glycerine. From pieces thus prepared most instructive specimens may be obtained. One would have supposed that in pneumonic lung there would be little difficulty in obtaining capillaries through the walls of which colourless blood corpuscles were in the act of passing—but although I have searched long and with great care, and in exceptionally good specimens, I have never

found an undoubted instance. I believe that the numerous bioplasts occupying the air-cells of the consolidated pulmonary tissue have resulted from the growth and multiplication of numberless minute particles of bioplasm which even poured out with the exudation. This view seemed to me the only one admissible after a careful consideration of the facts, and was advanced before the year 1860.

Cancer of the Lung.—In order to study a cancerous growth in the lung, the process of injection should be resorted to. The best plan would be to inject that part of the lung which was about to be invaded by the growth and the adjacent growth itself—with carmine fluid, and afterwards with Prussian blue fluid, as described in page 105. In this way the relation of the cancer to the vessels may be made out, and an opportunity might be afforded for demonstrating the precise way in which the morbid growths spread, causing the air-cells to collapse and waste, or invaded them, and occupied their cavity and led to the destruction of much of the neighbouring tissue. Different forms of cancer are described in Chapter XIX, and in the accompanying plates representations of several will be found.

Alimentary Canal.

366. Lips, Salivary Glands, Tongue, and parts about the Mouth.—Sections of the lips are to be prepared according to general plan already many times referred to. Injections of the loops of vessels are readily made, and the nerve fibres around these I have seen very distinctly in specimens injected first with carmine and afterwards with the Prussian blue fluid.

Salivary Glands.—The investigation of the salivary glands and pancreas scarcely requires any special remarks. The best idea of their structure is obtained by subjecting one of the smallest labial or buccal glands or Brunner's glands to examination. The ultimate follicles and epithelium are very easily demonstrated in specimens which have been soaked for some time in glycerine. It is often troublesome to trace the continuity of the duct with the follicles, in consequence of some of the latter covering its terminal portion. The ducts of the salivary glands and pancreas may sometimes be injected. In attempting this, it is advantageous to subject the organs to firm pressure for some time previously, as has been recommended in page 404, so that as much as possible of the fluid they contain may be absorbed, and thus the entrance of the injecting fluid favoured. Good sections may often be obtained from specimens which have been hardened in alcohol and soda. The arrangement of the capillaries is easily made out in specimens injected with Prussian blue or other transparent injection. If the vessels are injected with gelatine only, very instructive sections may be obtained. In such investigations, however, it is neces-

sary to make a vast number of sections, and examine them carefully, or the observer will not be able to form a correct idea of the structure of the gland.

In disease the structure of these organs becomes variously modified. Sometimes obstruction of the duct causes accumulation of the secretion and consequent dilatation of the ducts behind the point of obstruction. Small concretions sometimes obstruct the minute ducts, and the follicles in consequence become much altered.

Salivary calculi vary considerably in size, and are composed of phosphate and carbonate of lime with which a good deal of animal matter is incorporated, being deposited with the earthy salts. Small aggregations of epithelium, held together by mucus, probably form the nucleus of salivary calculi.

The nerve fibres distributed to the follicles of the salivary glands form networks of fine fibres upon and between the follicles. The dark bordered nerve fibres, supposed by Pflüger to be near their terminal distribution in the epithelial cells themselves really divide and subdivide into fine fibres which have a far more extended course than Pflüger supposed before they reach their ultimate ramifications and subdivisions upon the external surface of the follicle, and never enter or become connected with the gland cells at all.

Tongue.—The principal points of interest to be demonstrated in the tongue, are the different kinds of papillæ upon the dorsal surface, and the arrangement of the muscular fibres forming the substance of the organ. The papillæ of the tongue of the frog and other animals in which the mucous membrane is soft and moist, may be snipped off with a pair of fine scissors, and examined in glycerine or other solution.

The *epithelium* of the tongue is readily demonstrated. Portions may be scraped off with a knife and many hair-like processes from the papillæ in the central part of the tongue towards the back will be detached. These will be found to be entirely composed of epithelium.

The *vessels* of the papillæ of the fresh frog's tongue are seen more distinctly if the specimen be treated with a little dilute acetic acid or potash, but it is better to study their arrangement in a specimen the vessels of which have been injected with the Prussian blue solution and the bioplasts stained with carmine.

The arrangement of the capillaries in the palate, tongue, and all the parts about the mouth of the frog and other batrachia is very beautiful. It is not difficult to trace very fine nerve fibres to the summit of the papillæ of this class of animals, and I have described complex plexuses composed of excessively minute fibres, less than the $\frac{1}{100,000}$ of an inch in diameter, in my paper published in the "Phil. Trans." for 1864, vol. 155, page 443.

The tongue of the mouse and other small mammalian animals, will

afford clearer and more beautiful specimens than those of man and the larger animals. In order to demonstrate the general arrangement of the papillæ and the structures beneath, in the tongue of man and other mammalian animals, it is desirable to stain the bioplasm by injecting the vessels, first with the carmine and then with the Prussian blue injecting fluid according to the plan described in page 105. When this has been satisfactorily accomplished, small pieces about the eighth of an inch or a little more, in thickness, cut in different directions, are to be soaked for a long time in glycerine and chromic acid (p. 103) until they become hard enough, when very thin sections may be cut with a sharp knife. The sections should be made in different directions, and if the section knife be made to cut parallel to the filiform papillæ, even thin sections of these structures may sometimes be obtained. The sections must afterwards be rendered transparent by the addition of a drop of glycerine containing twenty or more drops of acetic acid to the ounce. In such sections the muscular fibres can often be traced quite to the sub-mucous tissue, where their tendons may be seen to become continuous with the white fibrous element of the corium. The form and arrangement of the muscles, and their mode of interlacement have been well described by Dr. Hyde Salter, in his article "Tongue," in the "Cyclopædia of Anatomy and Physiology." The reader is referred to this article, to a paper by Mr. Zaglas, "On the muscular structure of the Tongue of Man and certain of the Mammalia," in Goodsir's "Annals of Anatomy and Physiology," and to Todd and Bowman's "Physiology," for a description of the anatomy of the tongue.

367. Of Mucous Membrane generally.—Mucous membrane consists of one or more layers of epithelium, with a more or less firm subepithelial texture which is continuous with, and seems in many cases to shade into the submucous areolar tissue. Occasionally the tissue subjacent to the epithelium forms a transparent lamina (basement membrane). Such a structure is very distinct in the case of the uriniferous tubes the epithelial cells resting upon the basement membrane.

Beneath the youngest bioplasts of mucous membrane is a layer of areolar tissue (*submucous*, *sub-basement tissue*). Into this structure, muscular fibres, or their tendons, where these exist, are inserted, and in it ramify the vessels and nerves. The thickness of the mucous membrane and other characters of the several structures of which it is composed vary much in different localities. That of the mouth, especially at the back of the tongue, may be readily subjected to examination, and the different structures enumerated may be made out. It is desirable to inject the vessels with transparent injection, and cut thin sections through the mucous membrane and subjacent structures with a sharp knife. On the anatomy of mucous membrane, the student is strongly recommended to consult Mr. Bowman's article "Mucous Membrane,"

in the "Cyclopædia of Anatomy and Physiology," for although it was written many years ago, it contains very much that is still of interest and importance.

The epithelium of mucous membranes is very readily subjected to examination, and its character is found to vary much according to the locality from which it is taken. The chief varieties of epithelium, and the method of examining them have been referred to in p. 241. In order to obtain a specimen of epithelium from a mucous membrane, it is only necessary to scrape the surface gently with a knife, and place what has been removed upon a glass slide. After moistening it with a little water, syrup, or a mixture of glycerine and water, which does not cause the cells to become so turgid from endosmosis, the specimen may be placed in the microscope. To prevent the thin glass cover from pressing too strongly upon the specimen, one or two pieces of hair or thin hog's bristles may be inserted between the thin glass cover and the glass slide.

The epithelium of mucous membranes is liable to undergo various changes in character during the course of disease. When exposed to the air the soft, thin, and moist epithelial covering becomes converted into a firm, hard, almost cuticular investment. Such a change is often seen in the epithelium covering the *glans penis* when this is not protected by the prepuce. In cases of *prolapsus uteri* when the os is completely exposed, its soft mucous covering becomes harder and almost cuticular. Tubercles are not unfrequently found connected with the mucous membrane and skin of the genital organs as a consequence of *syphilis*. In structure they may be compared with *warts* which are developed upon cuticular surfaces in other parts of the body. There are, however, morbid alterations affecting the growth of the epithelial cells both of mucous membrane and skin far more serious than any of those above referred to. Sometimes from the mere irritation of a foreign body, a redundant and exceedingly irregular growth of coarse ill-formed epithelial cells takes place. The mass gradually increases in size, and if it be allowed to pursue its own course, may at length become so large and take up so much nutrient matter as to exhaust the patient's powers and cause his death. Some of these growths have received the name of *epithelial cancer*. These commence in the epithelium, but the structures beneath gradually become much involved in the succeeding pathological changes, until at last a growth results in which all the elements of the original texture have undergone changes which separate them widely from the corresponding normal anatomical elements.

368. Nerve Fibres distributed to Mucous Membrane.—The mucous membrane of the fauces and palate is abundantly supplied with nerves. Beneath the epithelium I have demonstrated marvellous plexuses, the arrangement of which I have figured. The finest fibres have been

represented as they appear under the $\frac{1}{25}$ of an inch object glass, in plate LX, fig. 2, as they appeared when magnified 1,700 diameters. The demonstration is very easily made in the case of the mucous membrane covering the convex surface of the epiglottis. Pieces of the epiglottis with the mucous membrane attached are stained with the carmine fluid and then properly hardened in glycerine and chromic acid. After three or four weeks have passed, excessively thin sections are removed one after the other parallel to the surface, placed in glycerine containing ten drops of acetic acid to the ounce, and after soaking for few days are to be examined in the usual way. The most promising pieces are to be well pressed by the thin glass, so as to make them much thinner, and, by frequently repeated taps upon the thin glass the tissues are to be slightly frayed out without the anatomical elements being at all smashed. The sections are then to be again introduced into the glycerine solution placed in a watch glass for a day or two, and examined and pressed as before. These operations will have to be repeated many times before the specimens become as clear as it is possible to make them. The pressure may be produced by a spring clip, but it will be found that the process is expedited by administering a number of sharp and firm taps upon different parts of the thin glass covering the section with a piece of wood cut to end in a blunt point, fig. 1, pl. LXII.

369. The sub-mucous areolar tissue may be very readily demonstrated by removing a small piece from the under surface of the mucous membrane with scissors, and tearing it up with needles. Beneath the hard cuticular mucous membrane of the æsophagus, there is an abundant layer of lax areolar tissue, which connects the lining membrane with the muscular coat beneath, and permits very great alteration in the form and capacity of the tube, during the passage of its contents. A small piece of this may be readily removed for examination. It will be found to consist of areolar tissue in which numerous vessels and bundles of nerve fibres are seen to ramify and divide and subdivide into smaller bundles as they pass to their distribution in the muscular tissue, or in the mucous membrane.

The thin layer of pale muscular fibres situated immediately beneath the delicate basement tissue upon which rests the columnar epithelium of the small intestine, was discovered by Brücke. The contractile fibre cells of which it is composed are arranged in two layers, one of which takes a circular and the other a longitudinal direction. This is termed the muscular layer of the mucous coat, to distinguish it from the muscular coat of the intestine which lies external to the sub-mucous tissue.

370. Villi.—Muscular Fibres.—Lacteals.—The villi are best shown by making a perpendicular section of the mucous membrane of the small intestines with a very sharp knife, taking care, if possible, to make them take one direction by allowing a stream of water to flow over them,

as referred to in describing the method of examining the papillæ of the skin in the next chapter.

After the vessels of the small intestine have been injected with the carmine fluid and afterwards with the Prussian blue fluid, small pieces of the mucous membrane are to be placed in acid glycerine (1 to 2 per cent.) until the tissues are found to be clear and fit for examination. The pieces are to be transferred to glycerine to which a few drops of the chromic acid glycerine (page 103) have been added. The pieces are so arranged that the villi *hang* downwards in which position they become hardened. From the intestine of any small animals thus prepared most beautiful preparations of the villi may be obtained.

The columnar epithelical cells, and the large flask-like cells, can be seen without any preparation, but from pieces of intestines preserved in glycerine I have obtained beautiful specimens.

The *muscular fibres* of the villi, seen first by Brücke, are to be demonstrated by washing off the epithelium, and treating them with a solution of acetic or nitric acid, composed of about four parts of water to one of acid, or by soaking in glycerine.

The *Lacteals* may sometimes be demonstrated in consequence of being filled with chyle at the time of death, but their arrangement may be very satisfactorily observed in the villi of a rat or mouse which has been fed upon a considerable quantity of fatty food for some time before death. The animal should be killed by dashing it suddenly on the floor, for unless death be instantaneous, the lacteals become emptied before they can be submitted to examination.

371. Thickening of the Sub-mucous Tissue.—Cancer of the Stomach, Intestine, and Rectum.—In certain morbid conditions, the sub-mucous tissue in this region is found as a hard, dense, somewhat transparent-looking layer, varying in different cases from the eighth or tenth of an inch to an inch or even more in thickness, and almost of a cartilaginous consistence. Thin sections may be very readily examined, and will, in many instances, be found to be composed of the original elements of the tissue, but coarser and more abundant than in health, with a certain proportion of granular matter, and a few badly-defined cells, the nature of which it is not easy to decide.

In many cases of the so-called "cancer of the pylorus," nothing more than the thickening of the submucous areolar tissue above referred to can be observed, and upon microscopical examination none of the cells characteristic of malignant growths can be detected. A similar condition is not unfrequently found affecting the sub-mucous tissue of the colon, and cæcum, and in other situations. It is important to distinguish this affection from the true *cancer*, as in its general appearance to the unaided eye it is found so closely to resemble scirrhus, although it is essentially different from this disease in a pathological point of

view. Microscopical examination generally enables us to decide the question.

In some I have proved that the thickening is almost or entirely confined to the muscular coat. In one of these sent to me by Dr. Hall, of Brighton, the walls of the pylorus were nearly an inch in thickness, and upon making a thin section of the firm, hard, fibrous-looking tissue and examining it under a quarter, it was found to be composed of coarse bands like those of unstriped muscle. "Archives of Medicine," No. III. The same remarks will in great measure apply to the so-called *cancer of the rectum*. Not a few of these cases really consist of thickening of the muscular coat of the bowel, and are not of a cancerous nature at all. The muscular tissue loses its contractile power, and not only does it not waste, but it actually undergoes increase. The nerves which are distributed to the fibres waste and degenerate into connective tissue. The altered muscular fibres are wider and thicker than those in health, but the material of which they are composed is harder, but quite as clear and transparent.

True cancer, however, is found in connection with different parts of the alimentary canal from the mouth to the anus. The meshes or areolæ of the areolar tissue between the mucous and muscular coats are much enlarged, and are occupied by cells having the general aspect of cancer cells. The fibres entering into the formation of the walls of these spaces are found to be more numerous and of increased thickness. To such growths the term *cancer* may be correctly applied. After a time the morbid changes involve the mucous membrane itself, and an irregular ulcerating surface which bleeds freely, is formed. In the case of an ulcer of this description, a little of the secretion from the surface, the surface itself, and the hardened tissue beneath, should be separately submitted to observation.

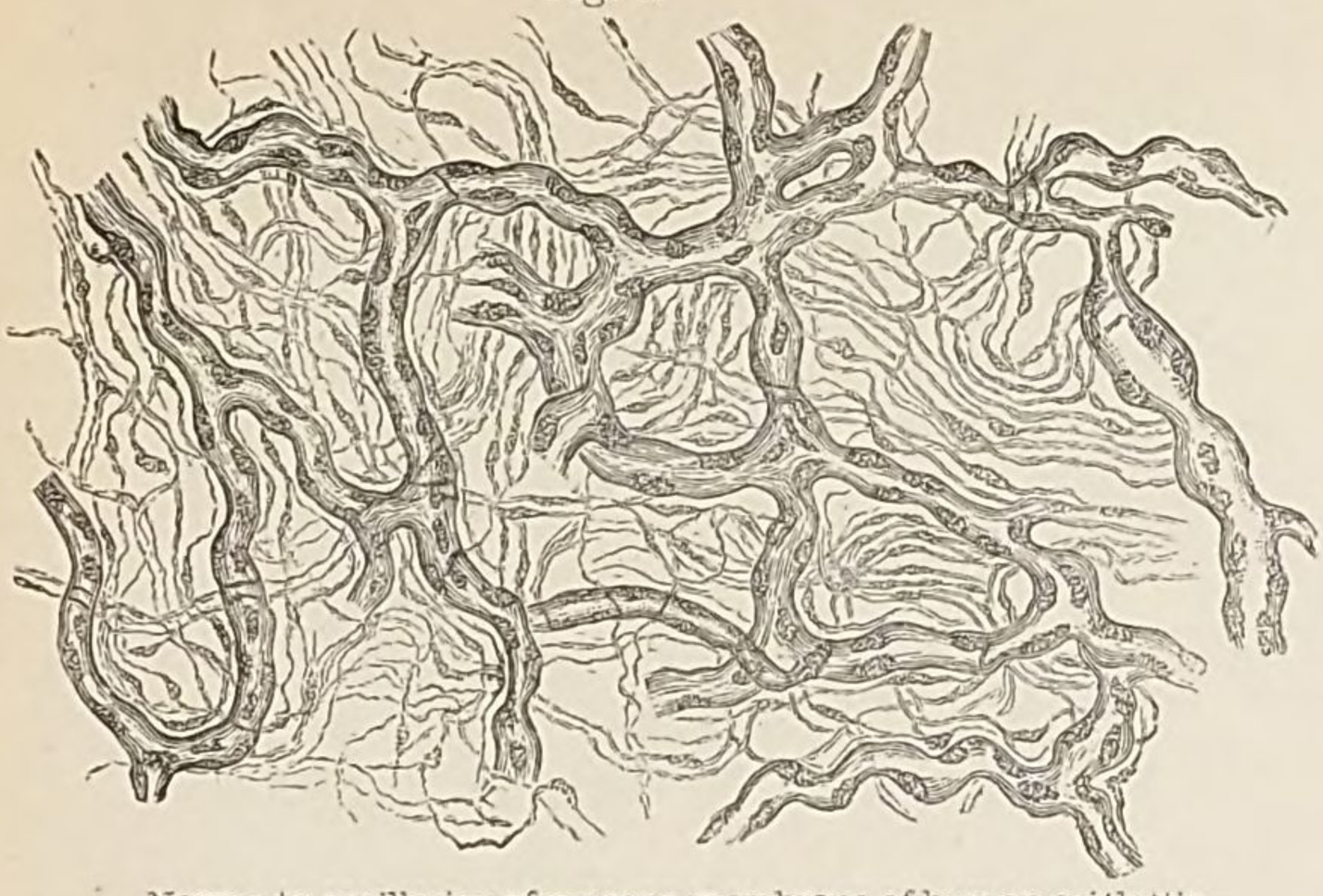
In the examination of these structures, thin sections entirely through the thickened mass should be obtained with the aid of a Valentin's knife. The section, after being slightly washed, may be subjected to examination with the usual precautions, or stained with carmine and preserved in glycerine.

The morbid changes occurring in the mucous membrane of the stomach in various cases of disease, have lately been investigated with great care by Dr. Handfield Jones, and more recently by Dr. Fenwick.*

372. Ulcers of the Stomach and Intestines.—The surface of ulcers of the intestine may be examined by scraping, or by cutting off small pieces with curved scissors. Sometimes the villi situated immediately

* "Medico-Chirurgical Transactions," vol. xxxvii, page 88. "Pathological and Clinical Observations respecting Morbid Conditions of the Stomach," by Dr. Handfield Jones. See also Dr. Fenwick's work on the Diseases of the Alimentary Canal.

Fig. 1.

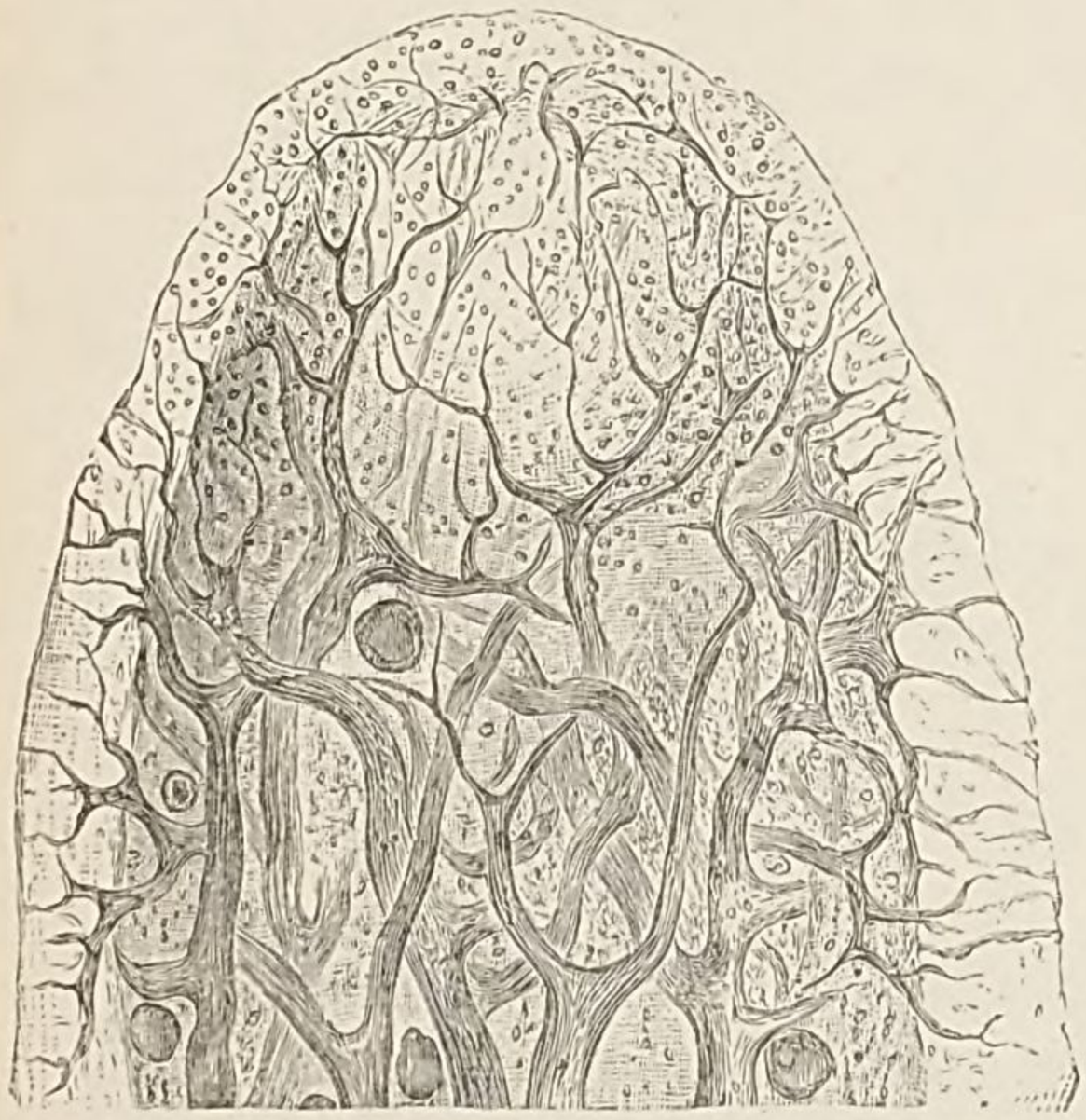


Nerves to capillaries of mucous membrane of human epiglottis. × 215. See also fig. 2, pl. LX. pp. 384, 396.

Fig. 2.

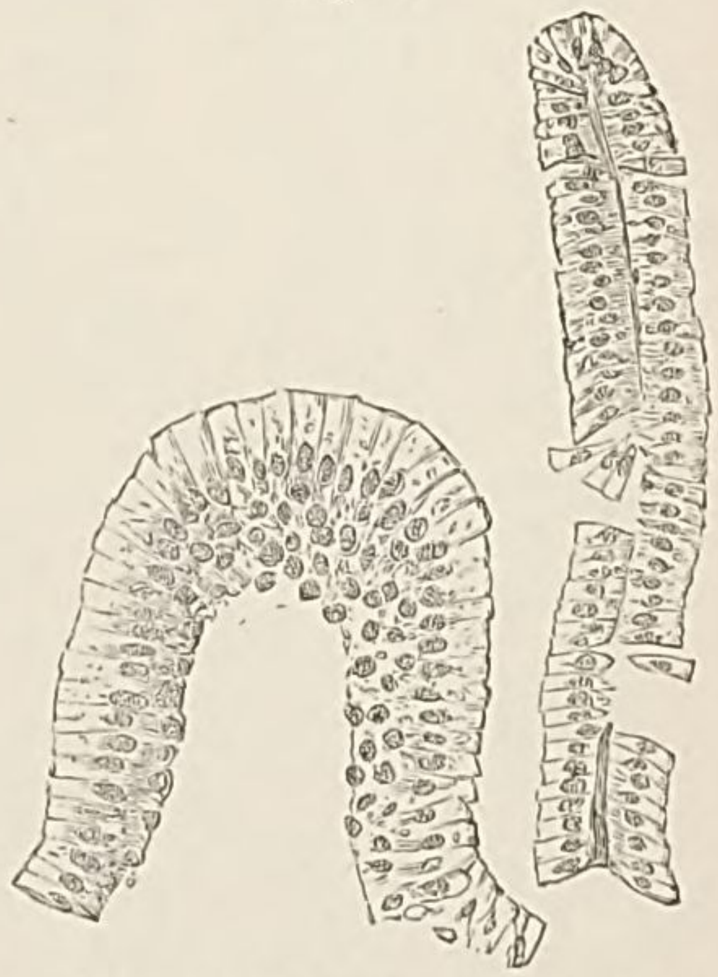
Epithelial sheath of a villus. × 130. p. 397.

Fig. 3.



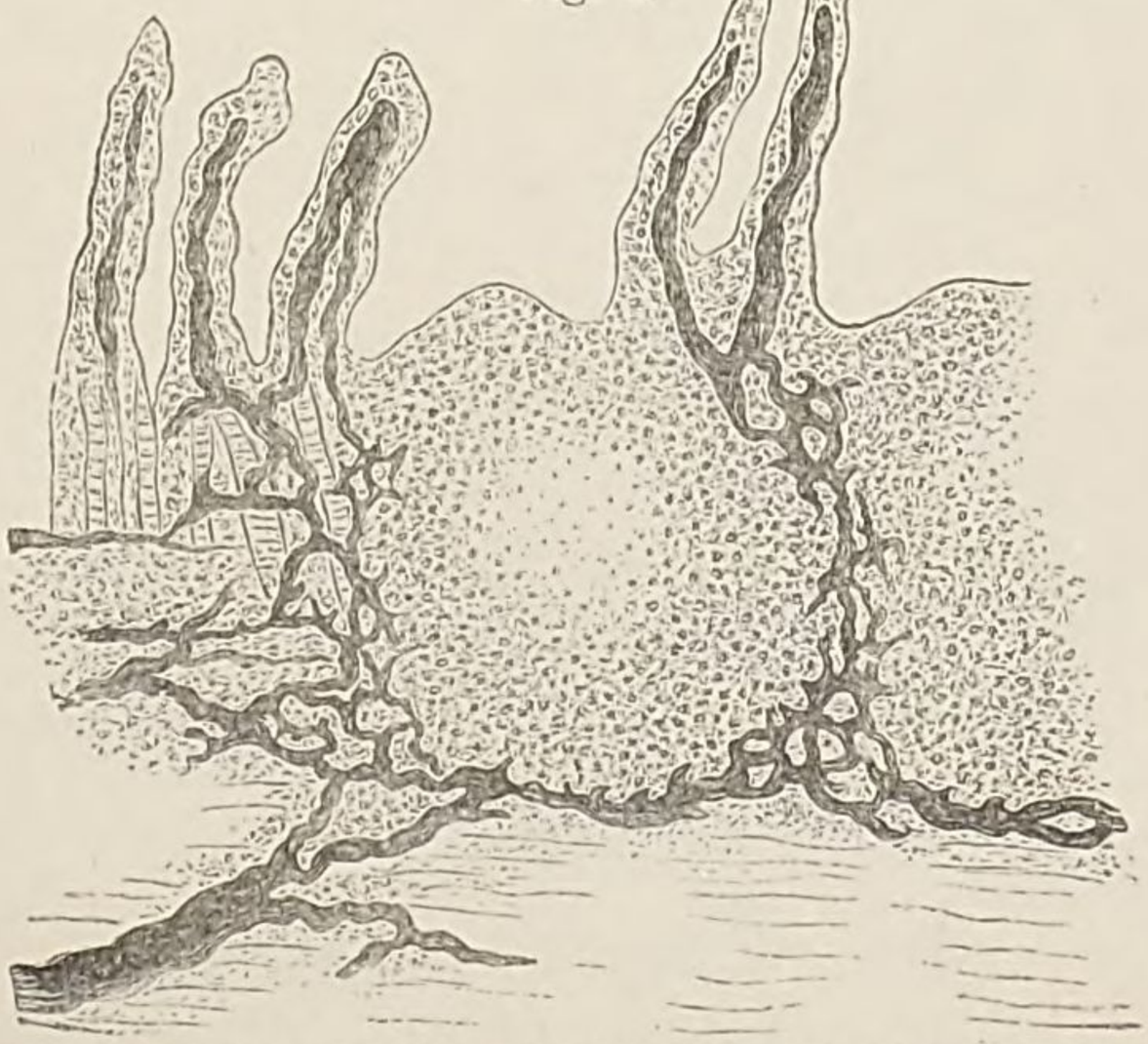
Summit of a villus from the jejunum of a patient who died of cholera, showing the shrunken and wasted vessels and the thickening of the tissue on the surface just beneath the epithelium p. 399.

Fig. 4.



Epithelial sheaths of villi, from a case of cholera. × 130. p. 397.

Fig. 5.



Villi of intestine with a portion of two glands of a Peyer's patch. p. 397. Lacteals injected. After FREY.

$\frac{1}{1000}$th of an Inch ⌊___⌋ × 215.

To face page 398.

around the ulcer, owing to the increased activity of the nutritive process which has gone on perhaps for a considerable time in the congested tissues, will be found to be very much increased in length. This remarkable change had taken place to a great extent in the villi situated around some ulcers of the small intestine of a patient, who died of fever some time ago in King's College Hospital. The villi round the margin of the ulcers were many of them three or four times as long as those in other parts of the intestine. Whenever ulcers of the intestinal canal are examined, we must always endeavour to ascertain if the ulcer has eaten into the muscular coat of the intestine, a point which can easily be decided by the presence or absence of unstriped muscular fibre in the tissue which forms the base of the ulcer.

There is yet much to be learnt with reference to the nature of various morbid changes affecting the mucous membrane of the intestinal canal, and especially the villi. The investigation will be best conducted after the vessels have been injected with Prussian blue fluid. A portion of intestine is readily injected if the pipe be inserted into a small trunk of an artery after all the divided vessels have been tied. Many of these may be gathered together and a ligature passed round at once, fig. 2, pl. IX, p. 104. On the remarkable changes in the villi in cases of cholera, see pl. LXII, fig. 3. A full description of the changes will be found in my work on "Disease Germs," 2nd Edition.

CHAPTER XVIII.

ON EXAMINING ORGANS IN HEALTH AND DISEASE: GLANDS: BRAIN AND SPINAL CORD: ORGANS OF THE SENSES, &c.—LIVER.—*Of injecting the Vessels, Ducts, and Lymphatics.—Of the Liver-cells. On investigating the Morbid Changes in the Liver.*—KIDNEY.—*Uriniferous Tubes.—Matrix.—Malpighian Tuft.*—SPLEEN.—*Thymus.—Thyroid.—Supra-renal Capsules.*—LYMPHATIC AND LACTEAL GLANDS.—SEROUS AND SYNOVIAL MEMBRANES.—BRAIN AND SPINAL CORD.—SYMPATHETIC GANGLIA AND NERVES. EXAMINATION OF THE SKIN AND ITS APPENDAGES.—*Cuticle.—Of making a Vertical Section of Skin. Sebaceous Glands.—Sweat Glands.—Hairs.—Molluscum.—Warts, Corns.*—NOSE.—EAR.—*Dr. Pritchard's Method of Examining the Cochlea.*—EYE.—*Of making Sections of the Sclerotic, Cornea, and Retina.—Examination of the Crystalline Lens.*—MALE AND FEMALE ORGANS OF GENERATION.—EMBRYONIC STRUCTURES, AND THE DEVELOPMENT OF ORGANS.

ORGANS OF SECRETION.

373. Liver.—General Examination.—The liver may be subjected to examination in many ways, and to demonstrate the various anatomical elements of its structure very different processes are required. If the cells alone are to be examined, a freshly-cut surface may be scraped with a sharp knife, and the matter thus removed covered with thin glass, and at once submitted to examination, or it may be necessary to add a drop of water or serum in the first instance. The appearance of a cell wall may be pretty distinct in water, but the sharp outline is due partly to the difference in refractive power of the water and the material of which the so-called cell is composed, and partly to the action of the water itself upon this. There is no membranous cell-wall as was formerly supposed. If the cells be placed in serum or glycerine, they appear perfectly solid, and no envelope whatever can be discovered, pl. LXV, fig. 1, p. 410. I have never been able to discern even the slightest interval between the supposed "cell wall" and the "contents" of the vesicle. We should certainly find such a space, under certain circumstances, if a cell wall actually existed.

In order to demonstrate the relation which the different anatomical elements bear to one another in this compact organ, we may cut a very

thin section by means of Valentin's knife, from the fresh liver; or still thinner sections may be taken from portions of liver which have been previously hardened in glycerine, alcohol, chromic acid, &c. The vessels of the liver may sometimes be demonstrated by washing the cells away from a thin section with a stream of water, and then treating it with a little dilute caustic soda. In specimens prepared in this way, however, the capillaries are often quite invisible. From the extreme tenuity of their walls in many cases, not a trace of them can be discovered; indeed the existence of the capillary wall can only be proved by filling the vessels with transparent injection in the first instance.

The investigation of the structure of the liver is somewhat difficult, owing to the numerous distinct tissues which compose the organ and the intimate connection of these with each other. I propose, in the first place, to refer to the mode of investigating the anatomy of the healthy liver; and, secondly, to indicate the methods applicable for ascertaining the nature of some of the morbid changes to which the organ is liable.

In the first place it is necessary to observe that the "lobules" so distinct in the pig's liver, in which each is inclosed in a capsule of fibrous tissue, are somewhat differently arranged in most other livers. In the human liver, and in that of most animals, although there is a mapping out of the entire organ into small elementary organs, or lobules, these are not separated from each other as in the pig, but the capillaries of one lobule communicate at various points with those of adjacent lobules. They are not separated by fibrous or other tissue, and no structure answering to the description given of Glisson's capsule, can be demonstrated in this situation. Great confusion with regard to the nature of the "lobule," has arisen from observers considering the pig's liver as the type to which others should be referred, whereas its arrangement is exceptional and totally different from the human and most mammalian livers. Physiologically the arrangement is much the same in all vertebrate livers, but anatomically great differences exist.

Of the Healthy Liver.—*Portal Vein.*—The general arrangement of the portal vein may be easily demonstrated by injecting one of the large trunks of this vessel. Any of the ordinary injecting materials may be used. It is desirable not to attempt to make a very complete injection, but to leave some of the capillaries, in the centre of the lobules, in an uninjected state, pl. LXIII, fig. 1.

Hepatic Vein.—The same process is applicable for demonstrating the arrangement of the branches of the hepatic vein. The injecting pipe may be placed in one of the branches exposed on the cut surface of the liver. The injection runs very readily, and upon examination it will be found that the capillaries in the centre of the lobules only are filled. Thin sections may be cut with Valentin's knife or with the double-edged scalpel; and it is desirable to take several thin sections

from the surface of the liver. The sections may be preserved in glycerine.

The portal vein may be injected in one part of a liver, and the hepatic vein in another part of the same piece. Sections of the latter, of course form the exact complement of those of the former. In the one, the central portion of the lobule has been injected, while in the other, the injection is confined to the vessels and capillaries at the circumference of the lobule. By injecting the portal and hepatic veins of a liver with different colours, these points may be shown in one preparation. Beautiful specimens of this kind may be prepared by injecting one vessel with carmine and the other with Prussian blue. See page 88, and Chapter VII.

Artery.—The arrangement of the artery is also shown by injection; the surface of the organ is supplied by an extensive arterial network, and the portal canals also contain a similar network. The coats of the ducts of the liver are so largely supplied with arterial blood, and the finest ducts are in such close relation with numerous small branches of the artery that it must be regarded as certain that some important chemical changes are effected in the bile by the oxygen of the arterial blood. The precise mode in which the arterial blood is poured into the veins has been a subject of great dispute, but I have many preparations which show that it passes into the portal capillaries near the circumference of the lobule as Kiernan long ago inferred ("Phil. Trans.," 1833), and not into those near the centre.

376. Of Injecting the Ducts of the Liver.—After death, the smallest ducts often contain a large amount of bile. By injecting water into the portal vein for some time, I found that a certain quantity permeated the capillaries and passed into the gall ducts. Thus the bile became diluted, and was forced out from the duct. Gradually in this manner the ducts were washed out, and every particle of bile was removed and made to flow out in the same direction as that in which it passed during life. Since the publication of my paper in the "Phil. Trans." for 1855, and memoir upon the anatomy of the liver, in which this mode of investigation was described,* some observers of high authority have expressed a doubt as to the possibility of forcing water through the vessels, to the extent advocated in my paper, without their rupture, and the destruction of the other structures. The plan which I first followed has been repeated several times, and in every instance the results which I previously arrived at have been confirmed. As the plan of proceeding may be of advantage in other investigations, I think it worth while to describe the operation somewhat at length, so that others may carry it out if they desire.

* "On the Anatomy of the Liver," illustrated with sixty-six photographs of the drawings, 1856.

Injection of the Liver with Water.—A large pig's liver within half an hour after its removal from the animal, was arranged as follows:—A piece of glass tube, the sharp edges of which had been removed, and one end a little enlarged in the blowpipe flame, was inserted into the *portal vein.* The vessel was tied round the tube with strong thread, all chance of slipping being prevented by the dilated extremity of the tube. A piece about four inches in length was inserted into the *hepatic vein* in the same manner. The liver was placed in a dish, over the head of which the tube inserted into the hepatic vein was allowed to project, in such a way that fluid flowing from it would be conveniently received in vessels placed near the stool upon which the dish was supported. A quantity of water at about the temperature of 100° Fahrenheit was placed in a vessel about four feet above the liver. The water from this reservoir was conducted to the portal vein by means of a glass syphon and India-rubber tube provided with a stopcock. Before connecting the flexible tube with the portal vein, some of the water was allowed to flow freely through it, and permitted to gravitate into the vein in such a manner as to allow all the air contained in that vessel to rise to the orifice of the tube before the connection was rendered complete. It is very necessary to prevent air from being driven into the capillaries; for if this should happen, rupture of the vessels and extravasation of the fluid will inevitably occur. The liver having been kept warm by the application of cloths dipped in hot water, the stopcock was turned so as to allow the water at 100° gradually to pass along the branches of the portal vein, and traverse the capillaries of the lobules. If such an arrangement be made we shall invariably notice that the entire organ soon swells to twice its size, while blood slowly trickles from the tube inserted into the hepatic vein. The blood soon becomes paler in consequence of its dilution with the water, the liver becomes tense, and the whole surface moist in consequence of the transudation of a little water; the small arteries are distended, the lymphatics are gorged, and the areolar tissue surrounding the vessels in the transverse fissure becomes puffy from the accumulation of water; bile passes along the duct, and the gall bladder becomes filled. Its contents may be forced out through the common duct by pressure, and it soon becomes re-filled, and this process may be repeated many times, the fluid which is removed containing less bile each succeeding time.

The water was allowed thus to wash out the vessels of the liver, and to permeate the ducts, for about four hours, and the fluid collected from the hepatic vein amounted to 344 ounces. The last portions which passed through were perfectly colourless, and contained no traces of sugar, which substance had been previously detected in considerable quantity. The liver was then removed, and injecting-pipes inserted into a branch of each of the following vessels distributed to different lobes:

—*portal vein*, *hepatic vein*, *hepatic artery*, and *duct*. A pipe should also be inserted into the branch of *portal vein*, distributed to the lobe in which the duct is to be injected. While the vessels are thus distended with water, branches are readily found, and the pipes can be inserted with ease. The liver was then wrapped up in soft cloths, small pieces of sponge being placed here and there, and subjected to considerable pressure during the next twenty-four hours, by being placed beneath a board loaded with about fifteen pounds.

It is desirable only to attempt the injection of the liver during cold weather, otherwise decomposition may have commenced before the fluid has been sufficiently absorbed to permit the introduction of the injection into the vessels.

After the water has been absorbed, the liver is very much reduced in size, and almost of a clayey consistence. The vessels are now quite empty, and ready to receive any injection which the observer may desire to introduce. As before stated, I have tried various kinds of the ordinary opaque injections, but although these may be forced in very satisfactorily, it is absolutely impossible that the arrangements of the duct can be made out, while the smallest branches can hardly be distinguished under these circumstances, as a higher power is required for their demonstration than can be conveniently applied to the examination of an opaque injection. For these, and several other reasons, I have used transparent injections, and give the preference to the Prussian blue solution.

Some of this injection was carefully forced into the several vessels, until the masses of liver were properly injected. It is desirable not to push the injection too far, as more is often to be learned from a partial injection than from one in which all the capillaries are completely filled.

We have, then, one lobe in which the *portal vein* is injected, another lobe injected from the *hepatic vein*, a third from the *artery*, and a fourth in which the injection has been forced into *the duct*. Of the three former, thin sections may be made after the lapse of a quarter of an hour, with a sharp double-edged scalpel, or with Valentin's knife. These may be gently washed on both surfaces, and immersed in glycerine. After having been allowed to soak in this fluid for half an hour, or longer, they may be placed in a cell and subjected to examination.

Before however the arrangement of the duct can be made out, a further operation is necessary. The injection forced into the duct will pass to the smallest branches, through which it will be conducted to the cell-containing network of the lobule. It will run amongst the cells and distend the tubes of this network to such an extent, that adjacent tubes will come into close contact—the capillary, which intervenes between them, being empty, or nearly so. If a section were made and examined, we should be able to make out nothing very definite; the duct could be

traced into the lobule and shown to be continuous with the injected portion, but the individual tubes could not be made out, or at least, only one or two here and there would be demonstrated. It is obvious, that if the capillaries were injected after the duct, this difficulty would cease, and the individual tubes of the cell-containing network would be separated by an injected capillary vessel. The lobe in which the duct has been injected is therefore to be placed in water slightly warm, and the portal vein injected with perfectly clear parchment-size. A pipe has already been inserted into this vessel. When the capillaries are quite filled, the pipe is closed with a cork, and the lobe placed in cold water until the size has completely set.* Thin sections may now be made in any direction, and as the tissue is very transparent, a small branch of the duct may often be followed for a very considerable distance. The sections should be preserved in glycerine. By comparing specimens from the different lobes which have been injected, the peculiar distribution of each vessel will be readily made out. A rabbit's liver is very easily injected; but in this case it is better to take one liver for each vessel, as the branches distributed to the different lobes are too small to receive the pipes.

After the pig's liver had been injected in the manner above described, thin sections were examined in the microscope, and with the aid of the neutral-tint glass reflector, their outline was traced upon transfer paper in the manner described in page 35, and drawings were made.† A drawing, copied from one of the preparations, showing the finest ducts of the pig's liver, and their expansion to form the tubes of the cell-containing net-work, is given in pl. XXXI, fig. 2, page 244. The blue injection is shown by the shading. The arrangement in the human liver is well seen in pl. LXIV.

377. On the arrangement of the Vessels of the Gall-bladder, Transverse Fissure, and Portal Canals.—The very peculiar arrangement of the vessels of the gall-bladder is referred to in my Monograph on the Liver, page 29. The only author who had previously noticed this beautiful disposition of the vessels, is Professor E. H. Weber,‡ but he makes no mention of a similar arrangement of the vessels in the transverse fissure, and in the portal canals§; and it is surprising that the very remarkable disposition of these vessels has not been represented by anatomists. As the arrangement of these vessels is very beautiful, and

* For this purpose it is better to employ a mixture of size and glycerine.—"How to Work with the Microscope."

† "Archives of Medicine," No. I, Plates I, II, III, and IV.

‡ "Annotationes Anatomicæ et Physiologicæ. Programmata Collecta Fasciculus II," page 225. "Berichte iüber die Verhandlungen der Königlich Sachsischen Gesellschaft zu Leipzig," No. III, 1850, s. 185.

§ The arrangement of the vessels and vasa aberrantia in the transverse fissure of the human liver is represented in fig. 25 of "The Anatomy of the Liver," and in fig. 1 of my paper in the "Phil. Trans." for 1855.

the preparation from which my drawings were copied tolerably perfect, I introduce the illustration in pl. LXIII. The gall-bladder, the transverse fissure, and the portal canals are, as is well known, abundantly supplied with arterial blood, especially in the neighbourhood of the ducts. In these localities there exists an arrangement which permits the free circulation of the blood through the arteries, and facilitates its return into branches of the portal vein. Each branch of artery is accompanied by two branches of the vein, and this arrangement exists even in the case of very small divisions. The small branches of the arteries anastomose very freely, in some cases forming five or six-sided spaces, so that an arterial network is formed. This is met with on the external surface of the gall-bladder, pl. LXIII, fig. 4, page 406, in the transverse fissure, and in the portal canals. The vessels composing this network are accompanied on either side by a branch of vein. These also form networks, and the two venous branches communicate freely with each other, by transverse branches which pass over or under the artery. The trunks of the veins and arteries are of course distinct, and the blood, as in other cases, passes through capillaries before it reaches the veins. The vessels described are from the eighth to the twentieth of an inch in diameter. Such an arrangement of double veins would facilitate the rapid return of the blood after it has passed through the arteries, and as each branch of the vein is as large as the artery, would permit the return of a larger quantity of blood by the veins than was transmitted by the arteries in a given time, in case the volume of blood should have been increased by the absorption of fluid (Archives of Medicine, No. II).

378. Liver Cells.—The hepatic cells even in a state of health, generally contain a few oil globules, which vary a good deal in size, but which are for the most part very minute. The oil globules lie amongst a granular formed material in which bile pigments can often be seen, and not unfrequently crystals of yellow biliary matter. The bioplasm usually nearly spherical in form, with its new centre (nucleus), is embedded in the formed material. See figs. 2, 3, pl. LXIII, fig. 1, pl. LXV.

In disease the cells may become wasted and shrunk; they may be filled with granular matter, or gorged with fat, or the fatty matter may have increased so enormously in quantity as to cause the obliteration of the cell form altogether, in which case a thin section of the liver will be found to present, under the microscope, an appearance not to be distinguished from ordinary fatty tissue. Not unfrequently the cells of the liver, especially those in the central part of the lobule, will be found to contain minute crystals or granules of red, reddish brown, and yellow colouring matter. The cells of a fatty liver contrast remarkably with the starved state of the cells of a scrofulous liver, or with the pale granular cells which are often met with in the livers of patients who

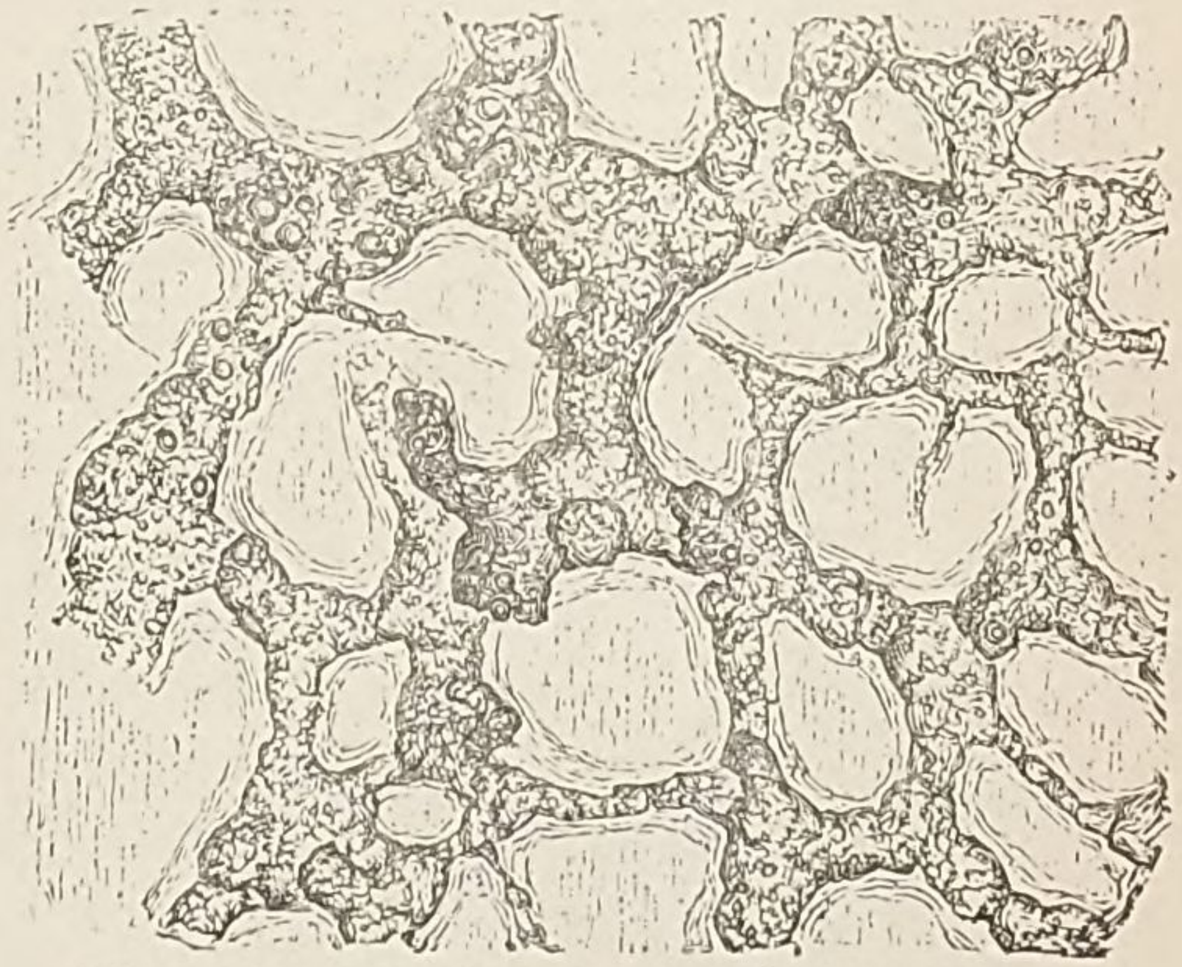
Fig. 1.

Portal vein.—Hepatic artery and Hepatic duct, well injected with different colours, in an interlobular fissure. Their divisions and sub-divisions on the surface of a lobule are well seen. From the human liver.
× p. 401.

Fig. 2.

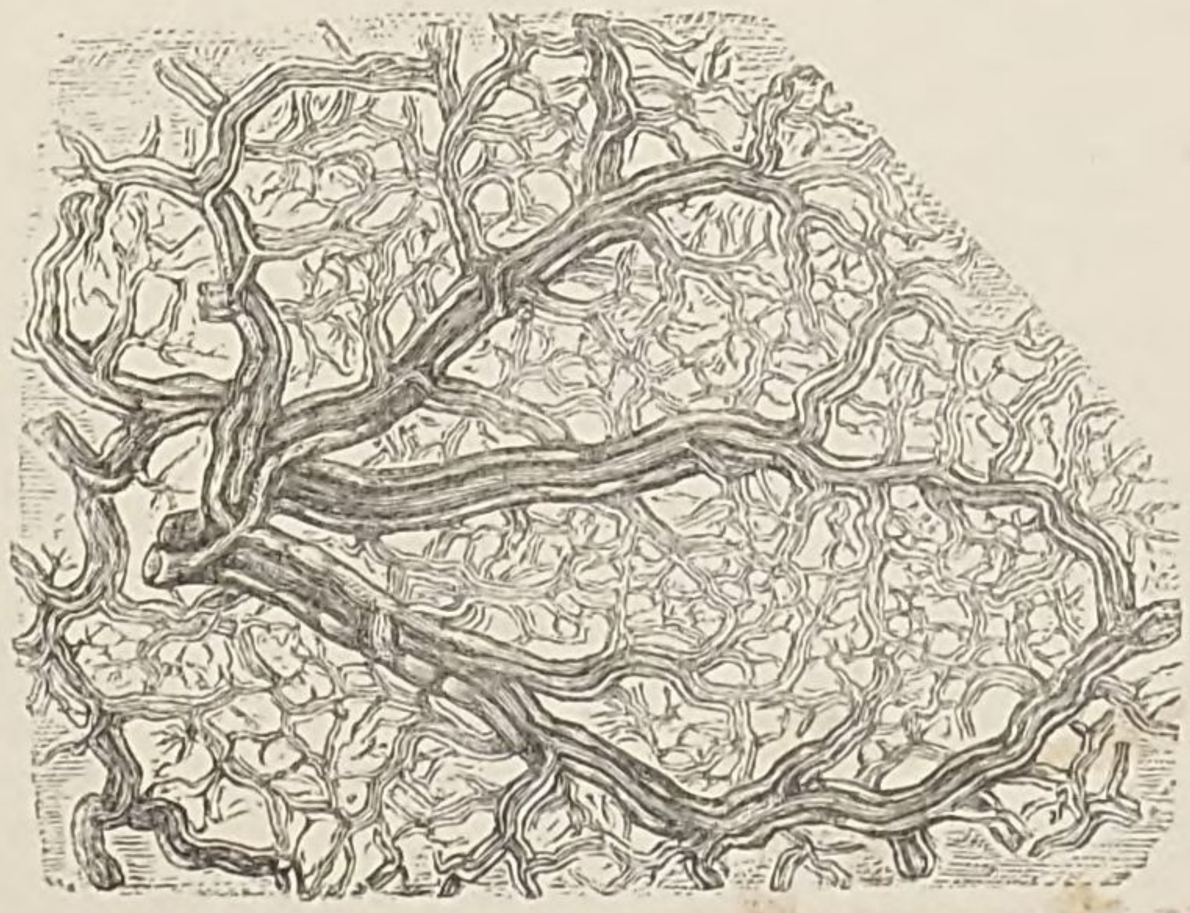
Capillary vessels and tubes of the cell-containing network. Healthy human liver. × 215. p. 407.

Fig. 3.



Cell-containing network near centre of lobule, somewhat shrunken. The capillaries were dilated to twice their normal diameter. × 215. p. 409.

Fig 4.



Ramification of artery and accompanying veins on the external surface of the human gall bladder, well injected. Veins white. Arteries between the veins, vermilion. Natural size. p. 405.

[To face page 106

1000th of an inch ,___, × 215.

have died of diabetes. Chemically, the amount of fat is found to vary much, and the balance shows the enormous increase in a more striking manner than the microscope. From a fatty liver which I analyzed many years ago, I obtained as much as 65·19 per cent. of fatty matter; and upon comparing this with the quantity obtained from a scrofulous liver, a remarkable difference was noticed, for in the latter only ·57 grs. per cent. of fatty matter were obtained.

The cells of the liver are somewhat irregularly arranged in the interior of delicate tubes so as to constitute a cell-containing network, the walls of which are so thin that in some instances only can they be demonstrated. In the frog, toad, and many more of the lower vertebrata, the continuity of the tubes of this network with the ducts can be proved beyond a doubt. In man and the higher vertebrates, the arrangement is sometimes to be very distinctly demonstrated in health, but in certain forms of disease in which contraction of the liver and thickening and condensation of the anatomical elements have been proceeding for some time, the appearances are such as render any other anatomical explanation of them inadmissible. Portions of the cell-containing network can be demonstrated clearly enough in sections of livers that have been hardened by being kept for a long time in syrup or glycerine or other medium containing a little chromic acid. The appearances are represented in figs. 2, 3, pl. LXIII, and many more will be found in my paper in the "Phil. Trans." for 1855, and in papers in my Archives, and elsewhere.

379. Of Injecting the Lymphatics of the Liver.—Many unsuccessful attempts had been made to inject the lymphatics of the liver, before the plan which ultimately succeeded was adopted. I had been able to force injection for some distance along the larger trunks in the opposite direction to that in which the valves opened, but I could not obtain satisfactory injections of the smallest vessels. The largest lymphatics in the portal canals are often injected by rupture of the coats of the duct, and by extravasation of the injection, as Mr. Kiernan remarked in his paper. Extravasation has many times occurred also in my own injections, but the injection always runs towards the transverse fissure, in the direction which the arrangement of the valves permits the fluid to pass readily, *and the smaller branches of lymphatics are never injected.*

The plan which I found successful was the following. An ox liver was thoroughly injected with warm water from the portal vein, as if the ducts were to be injected; gradually the organ became distended, and much fluid returned by the lymphatic vessels. Many large trunks running over the surface were fully distended with water. Into one of these swollen trunks a small pipe was inserted without much difficulty, and the vessel was tied round it. The whole organ was wrapped up in

cloths and subjected to considerable pressure for upwards of twenty-four hours. When the water had been absorbed, the lymphatic vessels were quite invisible, and it would have been impossible at this time to have found a lymphatic trunk into which a pipe could have been inserted. A little of the Prussian blue injecting fluid was now forced into the pipe from a small syringe. It passed for some distance along the trunk of the vessel, and by applying moderate pressure upon the surface of the large trunks from time to time, was made to pass the valves, and so was forced into the smaller branches. By using slight but gradually increased pressure, the trunks were so distended as to render the complete closure of the tubes by the valves impossible. In the course of half an hour or longer, an abundant network of lymphatics upon the surface had been fully injected, without any extravasation. It was now considered desirable to wait awhile, before introducing more fluid. After a few hours the injection was resumed until as much fluid had been forced in as could be introduced without running the risk of rupturing the vessels. I have tried the same plan with the human liver, but hitherto with very imperfect success, the trunks are much smaller, and their walls more delicate than those of the liver of the ox.

After the lymphatics had been injected as above described, thin pieces were removed for microscopical examination. Upon cutting thin sections from the surface, it was discovered that the injection had passed into many of the lymphatics of the portal canals, not only into the canals just beneath the capsule, but into some lying at the depth of an inch or an inch and a-half in the substance of the liver.

With the exception of my own, I know of no drawing or description of the smallest lymphatic vessels, either of the surface of the liver or of the portal canals and the appearances delineated in the drawings in Plate LXV, figs. 2 and 3, could only be obtained where a transparent injecting fluid had been employed, so that the specimen could be examined by transmitted light.

The network which lies partly in the substance of the fibrous capsule of the liver, and partly immediately beneath this structure, is fairly represented in Plate XIV, "Archives of Medicine," No. II. The smallest vessels have been injected, though in many situations not quite perfectly. There can, I think, be little doubt that the smallest branches form an intricate network. I have not been able to demonstrate the existence of blind extremities, and I do not think that lymphatic vessels commence in this manner. In the plate referred to, some very narrow branches are represented, many not being more than 1-2000th of an inch in diameter. In the preparation from which that drawing was taken, a network is seen in many places, and the branches which do not absolutely communicate are in many instances exactly opposite each other, a circumstance which renders it more probable that the

CONTINUITY OF THE DUCT, WITH THE CELL-CONTAINING NETWORK.

HUMAN LIVER.

A small terminal duct, A, of the human liver dividing into two branches, each of which is continuous with the tubes of the cell-containing network of two adjacent lobules. The "cells" lie in the tubes of this network, and injection has passed from the duct into the spaces between them and the walls of the tubes. × 215. p. 405.

1000th of an inch |___| × 215.

[To face page 40

tube in the interval is uninjected, than that there are cœcal tubes lying immediately opposite. The point at which the injection ceases is ragged, and of the same diameter as the rest of the tube, while if there were commencing blind extremities, they would be rounded, and probably a little wider than the rest of the tube. In many places the injection had accumulated in front of the valves, and had distended the tube very much.

In fig. 2, pl. LXV, from a portal canal, the injection is perfect, although doubtless the tubes are distended beyond their natural extent. Here, evidently, there is a network entirely surrounding the vessels and duct lying in the portal canals, and on either side of the large canal, smaller ones are observed to pass off. These also have their lymphatic vessels. No blind extremities can be found, and if they existed, I think at least a few would be distinguished in a part of the preparation where the injection is evidently very perfect. The walls of these finest lymphatics that were injected are so thin that minute particles of bioplasm might readily pass through them, but in no case have I seen indications of more minute tubes passing off laterally. If there had been such tubes even though less than the 1-50,000th of an inch in diameter, they would certainly have been injected. I have seen no appearances that justify me in accepting the conclusion that lymphatics commence in or communicate directly with the cavities or diverticula or prolongations of connective-tissue-corpuscles.

I have not been able to determine positively whether fine branches of lymphatic vessels enter the substance of the lobules from the portal canals, but many appearances I have seen render it probable that fine tubes pass a certain distance within the lobule, between the capillary vessels and the tubes of the cell containing network. In some of my specimens which lend support to this view there is no evidence of extravasation, and the appearance precisely accords with what one would expect to find if a lax network of fine lymphatic tubes existed in the lobule between the tubes of the capillary and cell-containing networks.

380. On Investigating the Nature of Morbid Changes occurring in the Liver.—To describe fully the methods of investigation which experience has proved most advantageous in rendering evident alterations in structure which have taken place in the liver in various forms of disease, would occupy too much space, but a few general observations in connection with the subject may possibly be useful to the student.

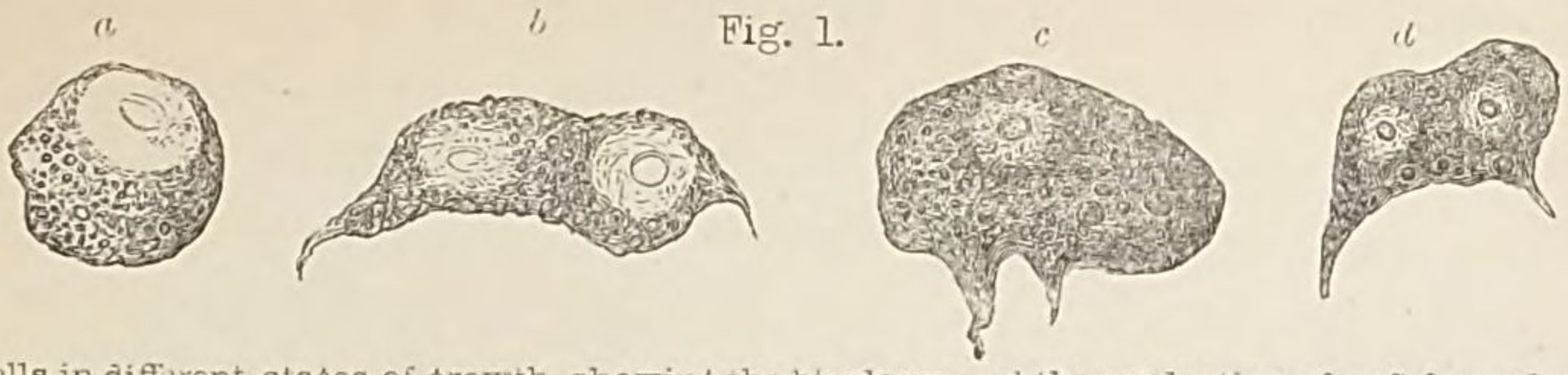
In investigating the morbid changes occurring in an organ having so complex and delicate a structure as the liver, it is of the first importance to demonstrate positively the precise locality of the changes. Although it would appear at first sight a simple matter to determine whether any given alteration was situated in the centre or at the

circumference of the lobule, it is often difficult to do so, for although the position of the artery and duct enable us to decide at once the intervals between the lobules, it is very difficult to distinguish these tubes unless they have been injected previously, and this proceeding cannot always be carried out in specimens removed at a post-mortem. The observer will find that the object is more easily gained by injecting a branch of the portal vein in one part of the liver, and one of the hepatic vein in another. In this way he will be able to distinguish the central from the circumferential parts of the lobule. If the Prussian blue fluid be used, it is not difficult to inject the vessels, and even very small pieces of liver may be injected in this way if contraction and condensation of the vessels and other structures have not proceeded too far. A very imperfect injection is all that is required to enable us to determine the exact situation of the particular morbid change. For instance, in some cases fatty matter accumulates in the centre, in others at the circumference of the lobules, and although the arrangement of the oil globules in the two cases is very different, and could be discerned by a practised eye, a student would be able to settle the point at once, and quite positively, if he took the trouble to inject a branch of the portal or hepatic vein. I need hardly remark that venous trunks of moderate size are readily distinguished from one another, and the portal vein is always accompanied by a branch of artery and duct, but these cannot always be seen. In many cases it is important to study and compare the character of the cells in different parts of the lobule and it is necessary to determine precisely the cells situated in the central part of the lobule and distinguish these from the cells at the circumference.

In some cases of disease, the tissues in the centre of the lobules waste, and when this wasting process has affected several adjacent lobules, an appearance as of interlobular fissures is produced, in the central part of the lobule. Again, the capillaries are prone to degenerate in some diseases, and their canals to be obliterated. This degeneration commences sometimes in the capillaries connected with the portal and sometimes in those opening into the hepatic vein—a point that cannot be easily determined until we decide which is the centre and which the circumference of the lobule.

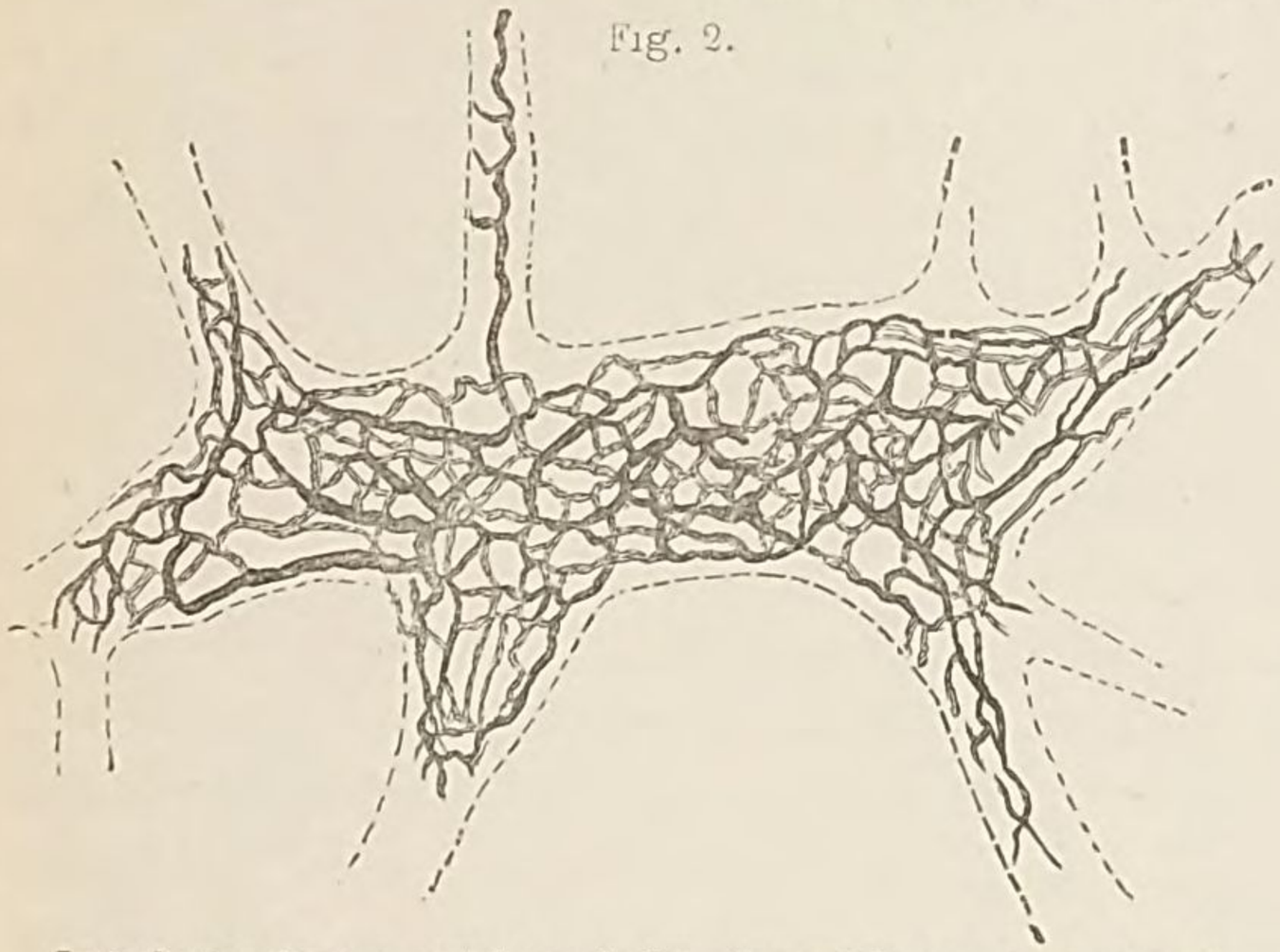
It has been held by many that *cirrhosis* consists essentially in the effusion of lymph in the interlobular fissures, and that by the contraction and subsequent conversion of this lymph into fibrous tissue the circulation in the lobule is impeded or entirely prevented. It is easily shown that this supposed lymph is abundantly supplied with vessels, and I have demonstrated branches of the duct in considerable number; indeed what is generally regarded as lymph consists really of the altered hepatic tissues at the circumference of the lobule. In every part of the so-called fibrous tissue the remains of the cell-containing network with

Fig. 1.



Liver Cells in different stages of growth, showing the bioplasm and the production of soft formed material, which becomes resolved into several different substances (fatty, amyloid, albuminous, and biliary matters.) Human subject × 700. p. 406.

Fig. 2.



Lymphatics from a portal canal of the liver of the ox. Several small branches pass into the smallest interlobular fissures. × 15. pp. 408. 423.

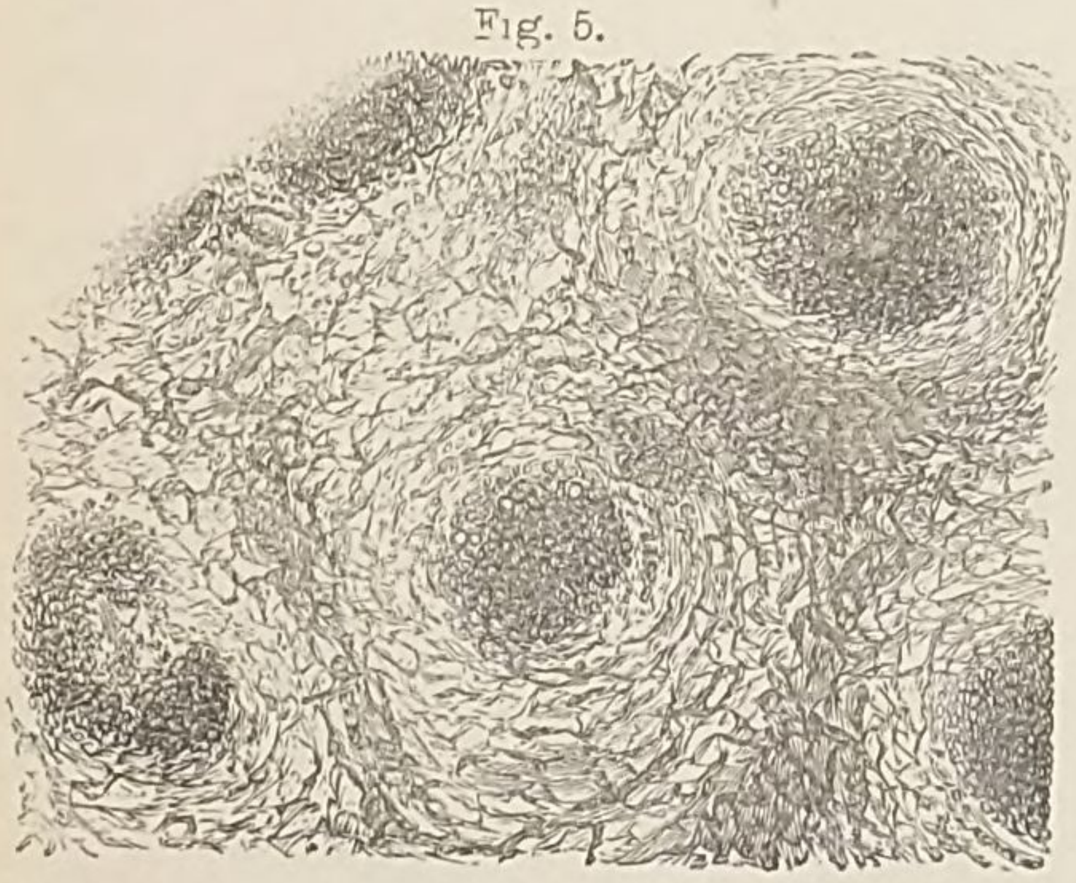
Fig. 3.

A portion of the network represented in Fig. 2. × 215. pp. 407, 423.

Fig. 4.

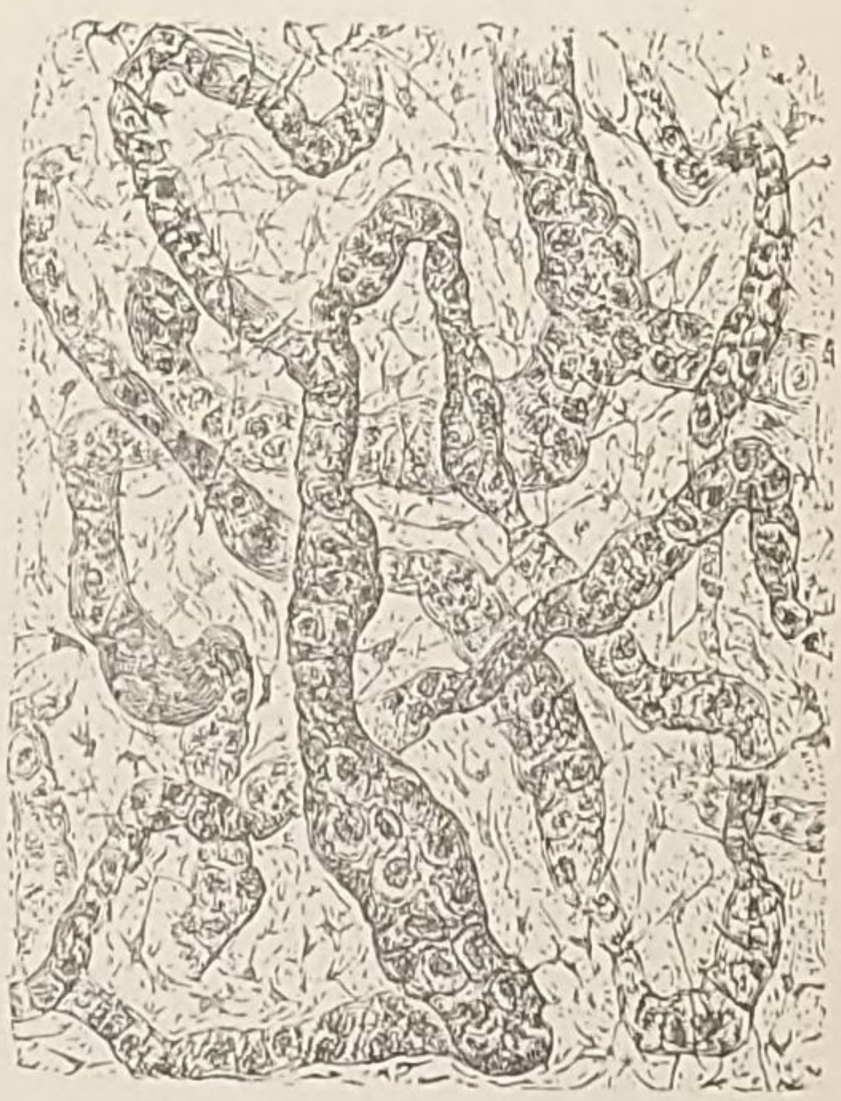
Three different portions of a cirrhotic liver, showing different degrees of change. *a*, Secreting structure wasting. *b*, Further stage. *c*, Secreting structure almost gone × 5. p. 410.

Fig. 5.



Three lobules, with a part of two others, showing the wasted and shrunken tubes of the cell containing network. Human liver cirrhosis. A part of Fig 4. × 20 p 410.

Fig. 6.



A portion of Fig 5, showing the wasted tubes and shrunken cells. × 215. p. 410.

1000th of an inch |—| × 215.

" " |————————| × 700.

To face page 410

shrunken, hardened and otherwise altered liver cells can be very distinctly seen. (" Archives," No. II.) Pl. LXV, figs. 4, 5, 6.

In certain conditions the walls of the capillaries appear to be much thickened, or an albuminous material is effused between them and the tubes of the cell-containing network,—or the thickening may, and probably does, affect both structures. In consequence, the distance between the cells and the blood becomes much increased. The selective agency and attracting property of the bioplasm operate through a greater distance than in health, and the changes in the blood induced by the formation of certain substances by the bioplasm of the cells, are imperfectly carried on. Such points as this cannot be demonstrated without great difficulty, and to ascertain the thickness of the capillary walls, it is necessary to make very careful injections.*

381. Kidney.—In the examination of the kidney, the epithelium and fragments of the tubes may be readily obtained by scraping the freshly cut surface. In this manner also Malpighian tufts may often be separated, but it is impossible to ascertain the relation of the different structures to each other, as by the process of scraping they are inevitably very much torn. A thin section in which this relation may be demonstrated, can be obtained either with the aid of a sharp thin-bladed knife, or more advantageously by the use of a Valentin's knife, by which means a section including both the cortical and medullary portion of the organ may be made. After washing the section very slightly, it may be placed with a drop of weak salt and water or weak glycerine between two pieces of glass, and examined in the microscope, first using a low power (an inch object-glass), by which the general arrangement of the tubes will be seen, and afterwards a quarter of an inch object glass, by the aid of which the different characters of the epithelium in the straight and convoluted portions of the tubes may be demonstrated.

But a far better plan is to inject the vessels of a kidney with carmine fluid and then with Prussian blue, according to the process described in Chapter VII. Small pieces are to be hardened in glycerine and chromic acid, and after two or three weeks thin sections are to be cut and immersed in acid glycerine, and afterwards mounted in glycerine.

Basement Membrane and Epithelium.—Just at the edge of the specimen, a portion of a tube stripped of epithelium, and exhibiting very distinctly the basement membrane, may often be observed, pl. LXVI, fig. 1. The thick polyhedral cells of glandular epithelium of the convoluted portion of the unriniferous tubes should be compared with the flat squamous cells which occupy the straight part. It will be found that in the latter the central channel is wider than in the former position, although the total diameter of the tube is less. This arises from the greater thickness of

* A few papers on the Morbid Anatomy of the Liver will be found in the numbers of the " Archives of Medicine."

the secreting epithelium in the convoluted portion. The epithelium of the convoluted part of the uriniferous tubes has been described by Dr. Isaacs, of New York, as tessellated.*

In looking at knuckles of tubes, an appearance is often produced as of a circumscribed cyst, arising, as Dr. Johnson long ago showed, from the bending of a tube as it twines in and out through the meshes of the capillaries, fig. 2, pl. LXVI. The cut ends of the tubes often appear thick and curled over, presenting in section the appearance of a thickened ring.

382. Matrix.—The appearance which has been described as resulting from the presence of a matrix may be seen very clearly in a section of the uninjected kidney of a mouse, or in that of many other rodents. Much discussion has arisen as regards the presence or absence of a fibrous matrix in the healthy human kidney, and the point is still considered by many as unsettled. The matrix was first described by Goodsir; and both Kölliker and Dr. Johnson have given drawings representing it very distinctly. In practice it is very difficult to say how much of the appearance is due to the presence of the walls of the tubes and capillary vessels, and how much to the existence of a structure (the so-called matrix) independent of these. Where the capillaries are injected with transparent injection, no *fibrous appearance* is to be detected; and I believe, at least in healthy kidneys, that the material resembling fibrous tissue, really consists of the walls of the tubes and the shrunken and compressed capillary parietes. I have been brought to this conclusion from examining a great number of specimens prepared in various ways. The only structure which exists besides these, is a small quantity of a clear transparent material, which is continuous with and may be said to connect the walls of the capillaries with those of the tubes. In healthy kidneys it is not easy to convince one's self even of the existence of this material. The fibrous appearance above alluded to may be observed if a thin section of the kidney be made with Valentin's knife and slightly washed in water. Such a question as this requires to be investigated in various ways before a definite conclusion can be arrived at. It is essentially necessary that the vessels

* Some confusion seems to have arisen in reference to the use of the term *tessellated*, which has been made by some authors synonymous with *polyhedral*. It seems better to restrict the term to those varieties of cells which are exceedingly thin and tile-like (if I may be permitted to use such an expression), and fitted to each other by their margins, after the manner of a tessellated pavement. If the cell was nearly as thick in one direction as in the other, the term would be inapplicable. The internal layer of choroidal epithelium is a good example of the tessellated variety, but the term clearly ought not to be applied to the epithelium of the liver and kidney. Dr. Isaacs, I think, wrongly describes the liver cells when he says that they "resemble tessellated epithelium," and he also considers the renal epithelium to be of the same character.—"Researches into the Structure and Physiology of the Kidney," by C. E. Isaacs, M.D., "Transactions of the New York Academy of Medicine," 1857.

should be slightly distended with transparent injection, otherwise their shrunken condition may easily be mistaken for a form of fibrous tissue. Figures 4 and 5, pl. LXVI, illustrate the different appearances in sections from the same kidney, the vessels in one case being injected with transparent size, in the other being left uninjected. In disease the fibrous appearance is more distinct, but I think it may be explained as well by referring it to the formation of a new tissue altogether, or to thickening and contraction of the walls of the vessels or tubes, as by attributing it to a hypertrophic thickening of the matrix. In some cases, in consequence of shrinking of the tubes and other changes, the meshes of the matrix appear more distinct than in health.

382. Vessels of the Malpighian Tuft.—Here and there, apparently upon the vessels of the Malpighian tuft, fig. 3 *d*, a few cell-like bodies are often seen. These have been described by some as epithelial cells upon the external surface of the vessel, but the researches of Mr. Bowman proved that the vessels are bare. The appearance of epithelium upon the surface of the vessels, is caused in part by the loops of capillaries being shrunken and collapsed, and in part by the bioplasts of the vessels. When distended with transparent injection the appearance is much altered and it is impossible under these circumstances to mistake the bioplasts of the vessels for epithelial cells or their nuclei.

Dr. Isaacs published a number of drawings to prove that epithelium existed on the vessels. In one of his drawings (pl. XXV), the diameter of the cells was greater than that of the uriniferous tube—which could hardly be the case, even if they had been much swollen by osmosis.* Although I have examined kidneys of various animals in different ways, and have tried the plan recommended by Dr. Isaacs, I have never been able to confirm his conclusions. His plan of proceeding was as follows :—Watery and ethereal solutions were injected into the ureter so as to burst the capsule, the Malpighian tuft having been slightly injected from the artery in the first instance. By soaking fine scrapings of the kidney, thus prepared, in water for a few days, the epithelial cells within the capsule were washed out, so that the space thus left between the tuft and capsule became filled with water which had soaked through the capsule. These processes seem simple enough, though I should think it a very difficult matter to inject fluid into the tubes so as to burst the capsule. I wish I had had an opportunity of seeing Dr. Isaacs' preparations, as I am compelled to confess that in all the attempts I have made, I have quite failed to demonstrate any approach to the appearances he has described and figured so distinctly ; neither have other methods of observation proved more successful in my hands.

Kidney of the Horse and other Animals.—The kidney of the horse is

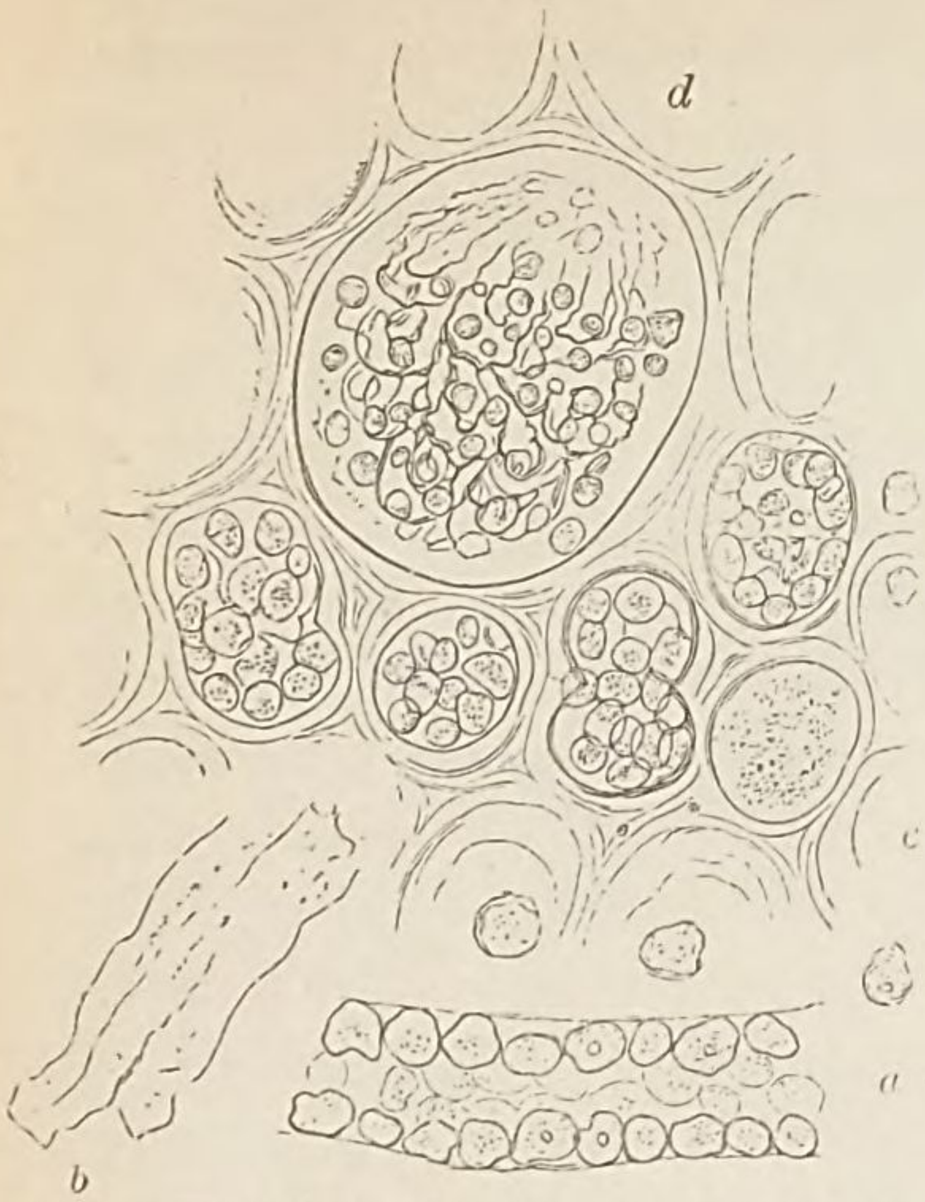
* "Structure and Physiology of the Kidney."—("Transactions of the New York Academy of Medicine," vol. i, 1857.)

well adapted for studying the minute anatomy of the Malpighian tufts, as in this animal they are unusually large, and, when injected, the arrangement of the vessels may be very distinctly demonstrated. A section of the straight portion of the tubes cut at right angles from a fresh kidney, will also show well the general anatomy of this part of the organ. In an injected kidney, such a section shows the outline of the tubes and their relation to the capillary vessels between. Sections should be taken from the cortical portion of the kidney, from the bases of the pyramids, and in two or three positions nearer their apices. Such specimens are very instructive, and afford an excellent idea of the general structure of the organ and of the relation of its different anatomical constituents to one another.

In thin sections of the kidneys of mice and other rodents, those appearances which have been considered to indicate the presence of the matrix are well displayed. All that is necessary is to cut a very thin section and wash it well previous to examination. The kidneys of rodent animals are very easily injected with the Prussian blue injecting fluid. The *afferent* and *efferent* vessels of the Malpighian body will also be made out in injecting specimens.

Notwithstanding the observations of Bowman originally published in the "Phil. Trans." for 1842, have been repeatedly confirmed by a number of the most careful observers who have worked since that period, we find new memoirs published for the purpose of disproving the main points which were then so conclusively demonstrated. One observer insists that the convoluted portions of the uriniferous tubes are not connected with the Malpighian body. Another asserts that there is no central cavity in the tube along which urine flows. A third maintains that the vessels of the Malpighian body are embedded in a thick epithelial layer. And as regards more minute points the differences are legion. It is of the utmost importance that broad questions of general anatomy should not be left in doubt and confusion. The view the student takes of course depends upon the text book which he has been advised to read. But every real student ought to appeal to nature and this at least in the case of the kidney he may do easily and successfully. I would strongly recommend the reader to proceed as directed in page 130, and carefully examine the thin part of the kidney of the newt. After a few trials he will be able to demonstrate all the important points of the anatomy of the uriniferous tube there referred to. But next let him inject a newt first with the carmine fluid (page 65) and afterwards with the Prussian blue fluid. Preparations of the same thin portion of the kidney are now to be made according to the directions given in page 105, and in other parts of Chapter VII. These specimens may be preserved permanently in glycerine, and may help to spread the truth and diffuse a knowledge of the real structure of the kidney.

Fig. 1.



Thin section of healthy human kidney slightly washed in water. *a*, convoluted part of uriniferous tube. *b*, tube stripped of its epithelium. *c*, outline of tube and crumpled capillaries having a fibrous appearance—the so-called "matrix" *d*, very small Malpighian body, the loops of vessels shrunk, showing bioplasts in their walls.
× 215. p. 411.

Fig. 2.

A part of the convoluted portion of an uriniferous tube from the newt's kidney, showing capillary vessels and nerve fibres, and the thickened basement membrane continuous in structure with the connective tissue. × 215. p. 412

Fig. 3.

Capillary vessels from the Malpighian tuft of the human kidney, which had been injected with dilute Prussian blue injection, showing nuclei connected with their walls. *a*. a few coils separated from the rest of the tuft. *b*, a portion of a loop somewhat compressed, showing the nuclei a little flattened *c*, tissue which connects the coils with each other, and thus preserves the globular form of the tuft even when entirely separated from the kidney. *d*, a small portion of a capillary pressed as much as possible, showing the thickness of the capillary wall at the point of reduplication, *e* × 403. p 412—13.

Fig. 4.

Fig. 5.

4, section of cortical portion of healthy kidney (human). Washed in water and examined in the same medium. The capillaries were not injected, and having collapsed and shrunk, exhibit appearance which is considered to depend on the matrix. 5, section of another part, in which the vessels were injected. The nuclei on their coats are seen, but no "fibrous matrix." × 100. p. 413.

$\frac{1}{1000}$ of an inch × 215

[To face page 414

384. Of the Bioplasts seen in a Section of Kidney.—Connected with the capillaries in every part of the kidney are minute bioplasts which were concerned in their formation and which play a more important part in connection with the functions discharged by the organ than is supposed. So near to one another are these bioplasts in the case of the walls of the vessels of the Malpighian body that when the vessels are uninjected the glomerulus appears to be made up almost entirely of them. Many are seen between the uriniferous tubes. Of these some belong to the vessels and some to the nerve fibres. Whether any ought to be regarded as belonging to the connective tissue is indeed very doubtful, seeing that it is very questionable whether that tissue itself exists. The greatly increased number present in many forms of disease may be accounted for as satisfactorily by the hypothesis that they originate from those in connection with capillaries and nerve fibres, as by attributing the increase to the growth and multiplication of bioplasts belonging to the supposed connective tissue. In certain forms of chronic nephritis and other conditions which are attributed to the development of connective tissue, or to the morbid increase of the normal connective, I believe the numerous bioplasts which exist in great numbers in the intervals between the tubes and between these and the vessels, result mainly from the division of the bioplasts of the tissues, and in very slight degree from the division and subdivision of colourless blood corpuscles and from the increase of the minute particles of bioplasm which have passed through the vascular walls. It is a fact that in many cases which are remarkable for the great number of intertubular bioplasts the capillaries are so much thickened that we cannot reasonably assume that colourless blood-corpuscles pass through in any number if at all. I believe that this process has really nothing to do with the pathological changes under consideration.

385. Microscopical Examination of the Kidney in Disease.—"Bright's Kidney."—This term has been employed to include all those morbid conditions of the kidney which are associated with albuminous urine. Of late years, however, certain important characters have been made out, which have enabled us to distinguish several morbid conditions of kidney, which result from special changes having taken place in that gland, and are essentially different from one another. Some physicians, nevertheless, still insist that the pathological states of kidney which have been described as distinct diseases, are really different stages of the same disease. It seems to me however, impossible, from a careful consideration of the facts at present known to come to the conclusion that the small contracted kidney so commonly met with in old drunkards, with its rough tuberculated surface, and diminished cortical portion, is but a different stage of a fatty kidney, or that it re-

sults from the occurrence of morbid changes, at all resembling those which end in the development of the large, smooth, and pale fatty kidney of young persons. Microscopically, as well as chemically, these forms of disease are distinguished by well-marked characters, while the history of the patient, the course of the morbid change, and its results, are totally different.

Dr. Johnson distinguishes "acute" and "chronic nephritis" from "fatty degeneration," and gives cases of three other forms of disease which have not been accurately described by any previous writer; these are "waxy degeneration" (corresponding to, and often found in conjunction with the so-called waxy degeneration of the liver), "non-desquamative disease," and "suppurative nephritis."—"Diseases of the Kidney," 1852.

Portions of diseased kidney are subjected to examination in the same manner as specimens of the healthy organ. Sometimes a thinner section will be required, in consequence of the opacity resulting from deposition of fatty matter, or an unusual quantity of epithelium, &c., within the tubes; or from thickening of the walls of the vessels, or the formation of a new material external to them. In the chronic nephritic kidney, the condition of the minute vessels should be especially taken notice of, as their walls are often much thickened, pl. LXI, page 388. The addition of a little acetic acid, or weak solution of caustic soda, often renders the vessels very distinct; and if the preparation be well washed in glycerine and water previous to examination, in order to remove the epithelium and granular matter from the tubes, the walls of the latter and the vessels will be more distinctly shown. Injected specimens of diseased kidneys should also be submitted to examination in all cases where it is desired to study the changes in the vessels.

The arterial thickening represented in fig. 1, pl. XLVII, page 422, probably depends upon degeneration and contraction of the tissues which entered into the formation of the wall of the healthy artery. At the time when the kidney was in a healthy state, the particular branch represented would have been, I believe, more than twice the diameter it now measures. It seems to me, therefore, that these changes belong to the category in which the degenerations of tissue would be included, rather than to that in which hypertrophic changes would find a place, but no doubt facts do exist which may be fairly considered to lend support to a different conclusion. I have described the changes which take place in my work on Kidney diseases, &c., from which several of the figures in the plates have been taken.

If the student desires to see kidney tubes the epithelium of which is filled with oil, he has only to examine the kidneys of a domestic cat, in which the convoluted portion of the tubes are nearly always loaded with oil, and oftentimes much oil is found in the Malpighian tuft. The

fatty matter is frequently so abundant as to cause the tubes, when examined by reflected light, to appear as if they had been injected with some white material.

Glands without Ducts.

386. Examination of the Spleen.—The investigation of the anatomy of the spleen is one of the most difficult researches the anatomist can undertake, and no one has yet succeeded in demonstrating satisfactorily the exact relation which the several anatomical elements bear to each other, and their development and mode of origin. The most favourable specimens are obtained from spleens which are tolerably hard when removed from the body; but at the same time it must be borne in mind that many of these firm hard spleens are not perfectly healthy.

The spleen of many of the lower animals, as of the rat, rabbit, cat, and many others, are firm and well adapted for investigation. The spleen, like other organs, may be hardened artificially, but owing to the large amount of blood and dark pulpy material which it contains, and which is usually rendered by hardening solutions not only hard but opaque, little is gained unless the sections are rendered transparent.

Different methods of investigation must be employed according to the structures to be examined. The various bioplasts in the spleen pulp are readily demonstrated by placing a small portion of the pulp on a slide and covering it with thin glass. It is important to look for large "compound cells," consisting of aggregations of blood corpuscles which are being disintegrated and reduced to a soluble state, and it is desirable to notice if any blood crystals are present, free or enclosed in the substance of corpuscles. The pulp should not be diluted with water, because physical changes in the anatomical elements would result. The characters of the different bioplasts under the influence of acetic acid and other chemical reagents must be noted. Blood crystals in the blood of the spleen were first noticed by Funke.

The most important constituents of the spleen, and the observation applies to the thyroid, lymphatic, mesenteric, and other glands belonging to this category, are bioplasts like colourless blood corpuscles, which vary greatly in number at different times even in the same individual, and which though growing and multiplying enormously in these glands are nevertheless found in nearly all tissues and in all parts of the body. They are connected with the absorption of nutrient matters rich in nitrogen, and are important agents in preserving the balance of the nutritive processes, and providing for an equable distribution of nutrient materials. In health numbers of these bioplasts soon undergo conversion into red blood corpuscles, or are otherwise disintegrated. If, however, conditions are not favourable to such further change, or to their death and disintegration, they may form threads of matter like fibrine,

which at length become converted into a form of connective tissue. In this way a delicate network of excessively fine fibres is often found, in the meshes of which many bioplasts are seen. In the course of months and years a great quantity of adventitious connective tissue may be developed in this way in the spleen. The texture of many other tissues and organs may be thus invaded and the discharge of their functions much impeded or completely interfered with. In this way many of the so-called "fibroid degenerations" are brought about, and certain forms of the process, as I venture to think incorrectly, called "fibrosis," are to be explained.

If it be desired to investigate the arrangement of the tissue of the spleen, it will be well to choose an organ which contains very little blood. I have obtained some instructive specimens by washing the blood vessels out, in the first instance, with glycerine ($\frac{1}{3}$) and water ($\frac{2}{3}$). When the blood is removed, the organ is placed in cloths and subjected to pressure in order to remove the solution; next, the vessels may be injected with glycerine, jelly, or some other transparent injecting material. I have some preparations in which exceedingly fine branches of the artery have been injected with the Prussian blue fluid, but I have not been able to demonstrate the mode of termination as determined by Mr. Gray. If the vein be injected, the appearance of extravasation is often produced. I have tried to inject the arteries with the Prussian blue fluid and the veins with plain size, and although this plan, or some modification of it, will in all probability be found to succeed ultimately, I have not been able to obtain specimens which conclusively demonstrate the most important points in connection with the structure of the spleen.

The *trabecular tissue* of the spleen is very readily examined, and its arrangement is well shown in specimens which have been injected with plain size, from which thin sections can be obtained without difficulty. Some portions should be treated with acetic acid, and the effects of other reagents should be ascertained. The muscular fibre cells are shown in portions of the tissue which have been immersed in diluted nitric acid. Dr. Billroth in his investigations, found great advantage from the use of sesquichloride of iron for the purpose of hardening the sections. (Beiträge zur vergleichenden Histologie der Milz, "Müller's Archiv.," 1857, page 88.) For a description of the anatomy of the spleen, the student is referred to Mr. Henry Gray's "Astley Cooper Prize Essay," Chapter XXXV of Todd and Bowman's "Physiology," and "Quain and Sharpey's Anatomy."

The connection between the appearance of a vast number of colourless corpuscles in the blood (Leucocythemia) and enlargement of the spleen, as well as the characters of the blood in this disease have been noticed in p. 263, but for full information upon the whole subject,

the reader is referred to Dr. Bennett's "Clinical Lectures on the Principles and Practice of Medicine," and to his work on "Leucocythemia."

387. Examination of the Thymus and Thyroid, &c.—These glands may be examined in the perfectly fresh state. Thin sections, obtained by a Valentin's knife, will afford the best specimens; the section will generally require washing slightly, in consequence of being covered with the softer and pulpy portion of the gland, which renders the arrangement of the tissue obscure. The glands may be hardened in a solution of chromic acid, in spirit, or in boiling water—but by these processes the cellular portion of the gland becomes somewhat modified. For making out the relation of the lobules and other structures to each other, hardened specimens are to be preferred. Very instructive specimens may be obtained from glands, the vessels of which have been injected according to the plan given in Chapter VII. In many of the smaller animals the thyroid gland is almost membranous, and this renders it very favourable for microscopical examination, especially in the case of injected specimens.

388. Suprarenal Capsules.—Until of late years the suprarenal capsules were very seldom examined even cursorily, and in the reports of many post-mortems, otherwise elaborately complete, the condition of these organs was not even mentioned. In the year 1855, Dr. Addison, of Guy's Hospital, published his memoir on "The Constitutional and Local Effects of Disease of the Suprarenal Capsules." The eleven cases there recorded, seemed to establish a relation between the deposition of brown colouring matter in the skin, and disease of the suprarenal capsules. Since that time, great attention has been paid to the state of these organs in all cases in which the colour of the skin has been observed to be at all darker than natural, and a great amount of very conflicting evidence has been obtained in this country and on the continent. Some observers have expressed themselves far more positively than Dr. Addison, in favour of the connection between the bronzing of the skin and disease of the suprarenal capsules; while others have brought forward a mass of evidence which to them appears perfectly conclusive against the existence of any such connection. The latter consider that the cases in which the bronzed condition of the skin is associated with disease of the suprarenal capsules, ought to be regarded merely as accidental, and they have taken great pains to collect examples of bronzed skin where the suprarenal capsules were healthy, and examples of disease of these organs where the colour of the skin was natural.

Now to many persons it would appear that to settle definitely a question of this kind could not be a very difficult matter. But it seems as if one set of men had been engaged in collecting evidence

while an opposite party was unworthily spending enormous labour and time in elaborate investigations for the sole purpose of upsetting the conclusions arrived at. In this way the problem was becoming extremely complex and more and more difficult to determine. Persons accustomed to scientific investigation, and really working with the sole object of ascertaining the truth, well know how very difficult it is to decide a question of this apparently simple character. On the one hand there is the danger of being led into error from drawing conclusions from insufficient data. On the other, the chance of losing perhaps for ever a valuable and important conclusion because the facts upon which it depends, appear at the time to shine very dimly through an overwhelming mass of doubtful and conflicting evidence. When such a question as this comes to be taken up by others, one finds that while one class of minds have a natural tendency to confirm the opinions of the first observer and perhaps involuntarily, but eagerly, clutch at any isolated circumstance which supports the original doctrine,—another class, possibly over cautious of receiving anything except upon the most incontrovertible evidence, plunges into the opposite error and, regarding as an accidental coincidence what is in truth a consequence, not only interferes with the spread of truth, but excites and encourages doubt in others. The difficulty is increased as the evidence on opposite sides accumulates, while a new element of discord is established by the record of a number of badly reported cases. It is only after the lapse of years that the evidence amassed can be thoroughly sifted and the truth at length discovered. As regards the present question we have perhaps reached the period when a positive decision might be arrived at.

It is important that all who are endeavouring to draw conclusions in this investigation, should consider the following points :—

1. That there are many cases on record of disease of the suprarenal capsules without bronzed skin, and that cases of bronzed skin have occurred without any disease of the capsules.

2. There is reason to believe that the term "bronzed skin" has been applied to very different appearances.

3. All observers who have reported cases, have not been sufficiently careful to describe minutely the state in which they found the suprarenal capsules. While some have spoken of these organs being in a morbid state upon the evidence of microscopical examination only, others have recorded cases only in which the changes were extensive and sufficiently evident to the most cursory investigation.

4. It should be noted that great differences are observed in the microscopical characters of these organs in health, or at least in the case of capsules which have been removed from the bodies of persons apparently in good health at the time of their death.

The subject of Addison's Disease will be found exhaustively treated in Dr. Headlam Greenhow's Croonian Lectures, at the College of Physicians for 1875. See "Medical Times" for 1875, vol. I.

The general form, size, weight, colour, and consistence of the healthy suprarenal capsules, are well known,* but it is not so easy to define the microscopical characters of these organs in a state of health. I think, generally, it is better not to attach much importance to the existence of a greater or less proportion of fatty matter than usual, in the cortical portion. Certainly only extreme alterations of this nature will justify the observer in stating the capsules were *diseased.* The microscopical characters should be accurately described and drawings given. In the cases brought forward by Dr. Addison, the alterations were so great that it was not necessary to resort to microscopical examination to discover them; but lately slight modifications in structure, only discoverable by minute observation, have been considered by some as sufficient to justify them in placing the case under the head of "diseased suprarenal capsules," although as yet we are ignorant of the nature and import of such alterations in structure, and indeed are unable to assert if they be morbid at all.

Although the drawings, figs. 2, 3, in plate LXVII, give a good idea of the general characters of the cortical portion of the suprarenal capsules in health, it is only right to state the fact that the proportion of fatty matter in capsules, which I believe to be healthy, varies greatly. The reader must therefore not put down as *diseased* every specimen which he finds differing in various particulars from my drawing.

The structure of the cortical and medullary portions of the suprarenal capsules is very different. The former consists of nearly parallel rows of cells containing many oil globules, lying in a meshwork of capillary vessels, and passing from the internal, towards the external surface of the organ. The latter is composed almost entirely of areolar or fibrous tissue, and nerve fibres with cells resembling ganglion cells,—and these are very abundant. Leydig considers that the yellow ganglion cells in the central part may be gradually traced into the cells of the cortex. Different opinions have been entertained with reference to the arrangement of the cells of the cortex. Simon thinks they lie in tubes; Kölliker, on the other hand, considers that there is no membrane, but that the cells merely lie in spaces; Ecker and Frey think that they are grouped together in oblong vesicles, but that these do not communicate with one another. It has not yet been satisfactorily de-

* They measure from an inch and a quarter to an inch and three-quarters in height, and about an inch and a quarter in width; their thickness is from two to three lines. The weight of each suprarenal capsule in the adult is from one to two drachms.—Quain's "Anatomy," vol. iii, page 329. Huschke estimates the weight at from 80 to 180 grains.

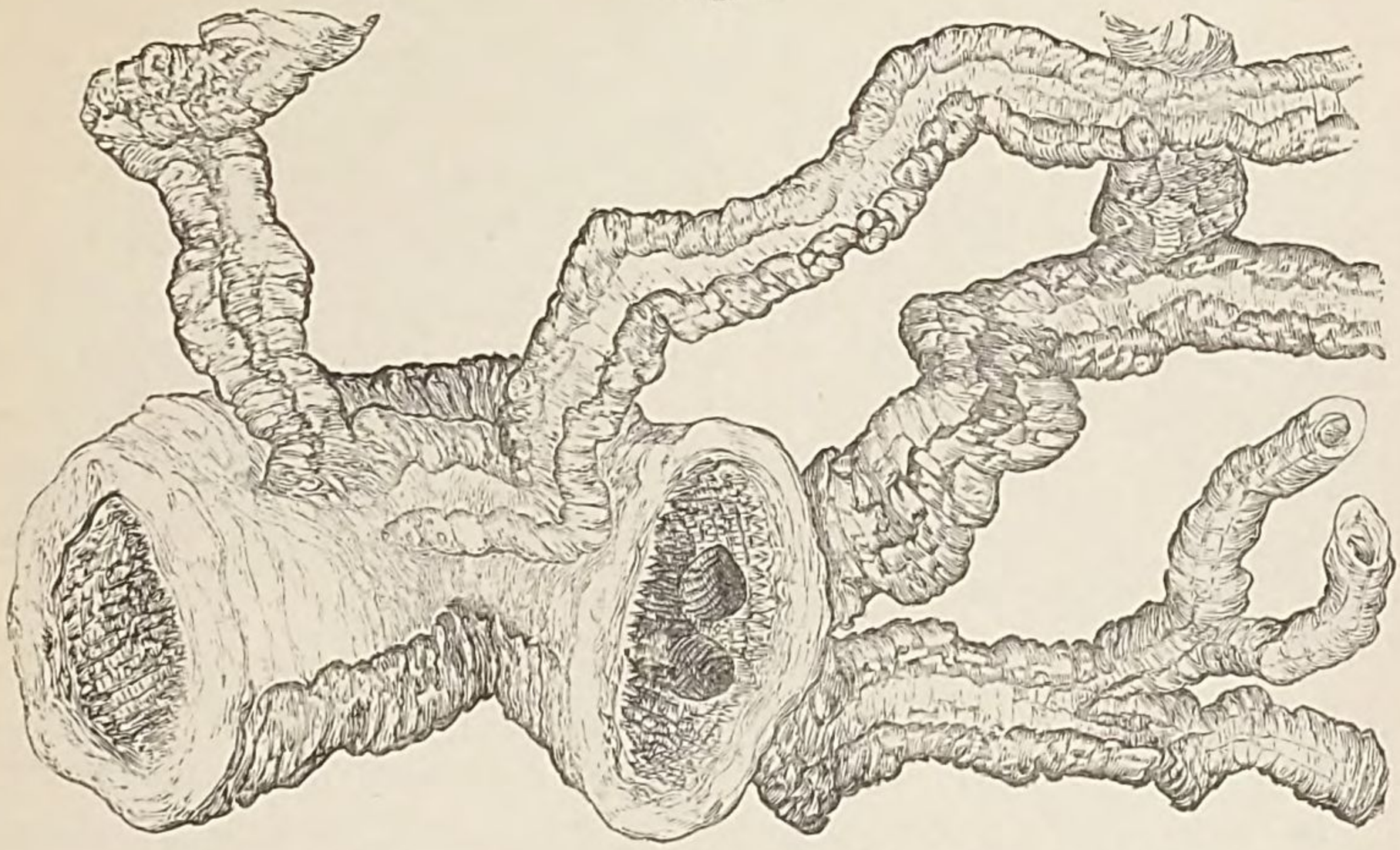
termined whether the cells are actually enclosed in a distinct tube, or merely take a linear arrangement.

389. Examination of Lymphatic and Lacteal Glands.—In the examination of these glands, a little of the thick fluid which is poured out from a freshly-cut surface, should be placed between glasses, without the addition of water or any other medium, and examined in the microscope. The student should make himself very familiar with the character of the bioplasts obtained from lymphatic glands, and should carefully observe their behaviour upon the addition of acetic acid, dilute solution of caustic soda and other reagents, and he should compare the changes which take place, with those which occur when the white corpuscles of the spleen, and white blood corpuscles are treated in a similar manner. The resemblance of these bodies to pus-globules should also be borne in mind, and the reaction of the latter should be very carefully studied. The minute structure of lymphatic glands is an exceedingly difficult subject to investigate. Many years ago my friend, Professor Donders, of Utrecht, gave me a beautiful specimen of a mesenteric gland of the dog injected with vermilion, which well showed the dense network of lymphatic vessels, of which the substance of the gland is in great part composed. The student should endeavour to make preparations in which the lymphatics or lacteals have been injected with one colour and the blood vessels which supply the gland with another.

390. Examination of Lymphatic Vessels.—The arrangement of lymphatic vessels may be shown by artificial injection. In consequence, however, of the valves opening *towards* the large trunks and preventing the flow of fluid from these towards the smaller vessels, there is considerable difficulty in making good artificial injections. Occasionally when blood-vessels are injected, rupture occurs and extravasation into the lymphatic vessels takes place. The lymphatics are often injected in attempts to inject the ducts of the liver, and sometimes by making a small opening into the areolar tissue beneath the skin or mucous membrane, the injection enters the lymphatics. In all these cases, however, only the trunks of the vessels can be displayed, and injection seldom passes into the smallest tubes. Formerly the lymphatics were injected with mercury, but although this process was well adapted for displaying the course of the larger trunks, the great distension produced rendered it impossible to ascertain the arrangement of the smaller vessels, even supposing them to be injected,—but the mercury seldom penetrated into very small branches. Opaque injections are not adapted for showing the smaller lymphatic vessels, especially as the large trunks are always much distended. The use of transparent injections affords more satisfactory results, and with the Prussian blue fluid I have prepared some very good specimens. Many attempts will probably be unsuccessful, but sometimes sufficient of the

DISEASED ARTERY, KIDNEY—SUPRA-RENAL CAPSULES.

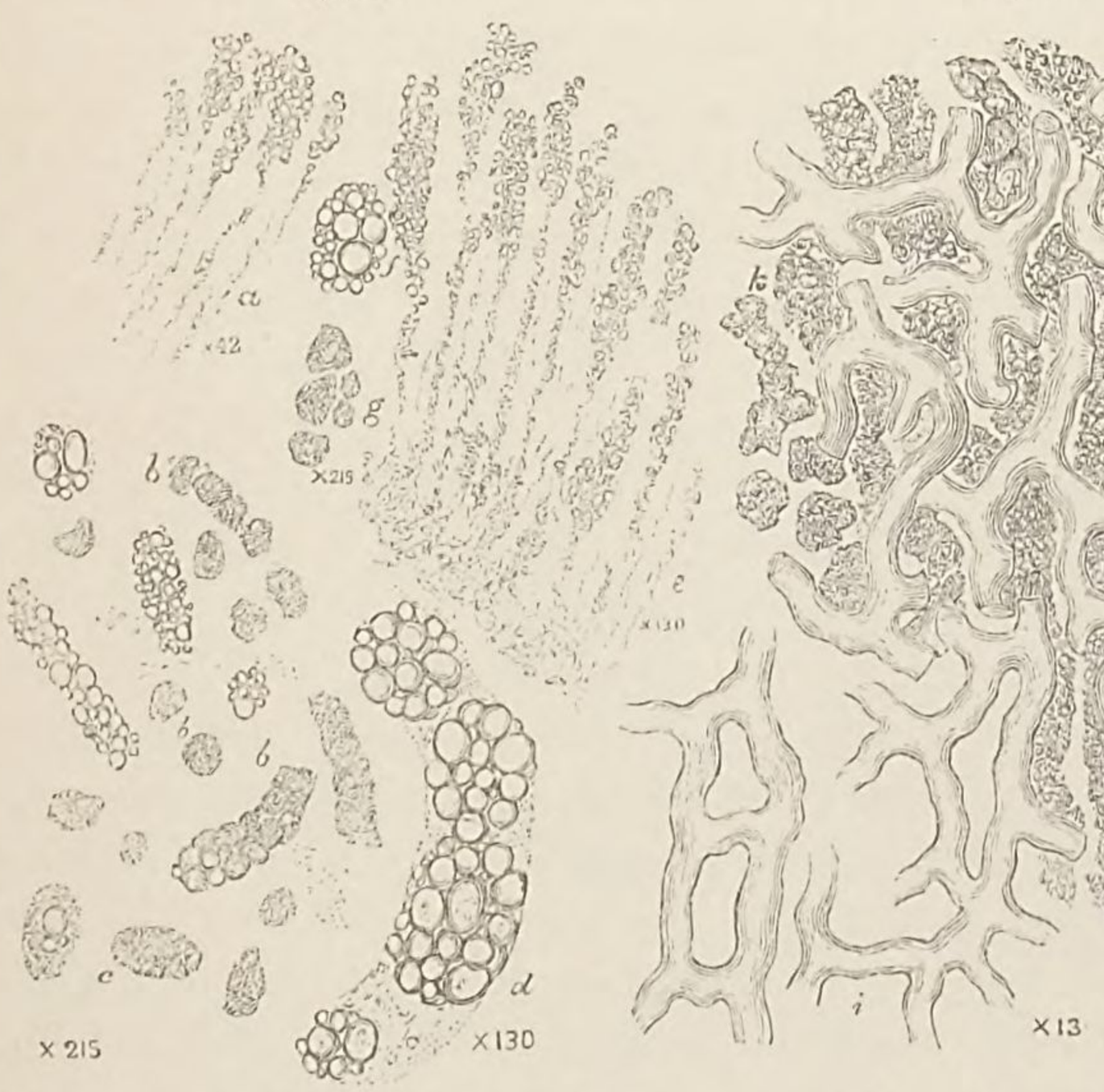
Fig. 1.



Branches of artery from a fatty contracting kidney, showing complete degeneration of the muscular fibre cells and deposition of glistening albuminous material. The walls of the artery have probably long lost contractile power, and are now converted into rigid inelastic tubes, the inner surface of which is uneven, with great irregularity in calibre. × 215. p. 416.

Fig. 2.

Fig. 3.



Structure of the healthy supra-renal capsule. Man. Fig. 1. *a*, from the cortical portion. *b*, small coloured cells. *c*, oval cells or collections of granules. do. *d*, cell-like collections of large oil globules. *e*, inner portion of cortex and outer part of medulla. *f*, oval collection of oil globules, from the part of the tube opposite to which it is placed. *g*, from the innermost part of the cortex. × 215 Fig 3. Vessels of a healthy supra-renal body injected, the lines of cell-like bodies, *h*, also shown. p. 421.

1000th of an inch ⌊___⌋ × 215.

[To face page 422

fluid may be made to pass the valves to inject the smallest branches, when the pipe has been inserted into a large trunk. If the vessels of the part be injected with water, much of the fluid will return by the lymphatics, the trunks of which become distended and rendered so large that a pipe may be very easily inserted into one of them. After a time, the water may be absorbed by cloths, and the injection forced into the empty vessels. In some instances it runs freely, and beautiful injections, even of the smallest branches, may be readily obtained. This plan, however, does not succeed in every case in which it is tried. Occasionally the lymphatics are distended with granular matter and oil globules to so great an extent, that their arrangement can be made out without any preparation. Lymphatics are seen in pl. LXV, figs. 2, 3, p. 410.

In fortunate injections of some of the finer ramifications of lymphatic vessels, appearances are seen which almost lead to the inference that valvular slits exist which readily permit fluid and minute particles of bioplasm, or other matter to pass into the lymphatics, but prevent the flow in the opposite direction from the interior of the vessel.

391. Examination of Serous and Synovial Membranes.—Serous membranes may be examined according to the general directions previously given. It will sometimes be found difficult to demonstrate the delicate cells upon their surface, and fresh specimens only should be studied for this purpose.

A small portion of the peritoneum of a mouse or other small animal, will be found to display well the fibres of the sub-basement tissue, and often vessels and nerves may be seen beautifully distinct in this situation. The greater part of the thickness of serous membranes is made up of condensed areolar tissue, in which the yellow fibrous element is very abundant. This areolar tissue becomes less dense at a greater distance from the surface, and often contains fat cells like the subcutaneous areolar tissue. In disease, the epithelium often increases very much in quantity; and in old cases of ascites or pleurisy, it is not uncommon to find the serous membrane completely altered in structure, its surface being covered by a tolerably thick layer of spherical or oval bioplasts. Frequently the fluid contained in the cavity is rendered turbid by the presence of a great number of bioplasts of a similar character.

In order to examine the distribution of the vessels in synovial membranes, an injected specimen is necessary. The fringe-like processes which project into many of the joints are highly vascular, and a well-injected specimen forms a beautiful object. The surface in the recent state is covered with large bioplasts more or less globular in form.

There are few things more worthy of study than the serous pericardium, or pleura, or peritoneum, in a state of acute inflammation. In the first the student has an opportunity of investigating the various changes which occur in that most important pathological change which

begins in increased nutrition of bioplasts of the tissue and exuded from the blood and ends in the development of a low form of fibrous tissue, and the last which, beginning in the same, ends not only in the formation of pus, but a form of pus which possessing specific poisonous properties is capable of producing the most terrible pathological phenomena in an organism into which it is inoculated.

392. Examination of the Dura Mater, Pia Mater, and Brain.—The examination of the dura mater and arachnoid may be conducted according to several different methods. Very small pieces of the fresh membrane may be removed, carefully torn with needles upon a glass slide, moistened with water, and covered with thin glass. In order to demonstrate the vessels and nerves of the dura mater, the membrane must be properly prepared by injecting the vessels first with carmine fluid and afterwards with Prussian blue. The nerves can be demonstrated in very thin pieces of the membrane which have been long soaked in glycerine. But for this investigation the student will find it desirable to examine the dura mater of small animals.

The gritty substances (brain sand) in the pineal body and in other parts, and the round starch-like bodies, which are not unfrequently met with in various parts of the brain known as *corpora amylacea*, may be separated from the brain substance by washing in a glass of water. The bodies in question will sink to the bottom; the supernatant fluid may then be poured off, and replaced by fresh water. After this process has been repeated a few times, the particles will become quite clean. They may then be examined in water, and tested with appropriate reagents. These may be preserved in aqueous fluid, or dried and mounted in Canada balsam.

The vessels of the brain may be readily examined if the white or grey cerebral matter be first removed from the pia mater, by washing with water. The addition of a little very dilute caustic soda renders the outline more distinct. But in order to examine the vessels successfully they must be injected with carmine and Prussian blue fluid, according to the plan laid down in p. 105. Very beautiful and highly instructive specimens may be thus prepared, and they should be mounted in strong glycerine, and examined under the highest powers, figs. 2, 3, pl. LXVIII.

The investigation of the anatomy of the central organs of the nervous system is difficult, and it is not easy to lay down principles for the student's guidance in all cases. Very much yet remains to be discovered with reference to the chemical solutions adapted to render the anatomical elements of these tissues distinct. There can be no doubt that methods of investigation will be discovered which will enable us to demonstrate satisfactorily the relation to one another of the delicate structures which make up the nervous system. The observations made in pp. 45–50, should be referred to, and the student should try for himself a number

of fluids of different composition. I cannot too strongly recommend the plan invented by Mr. Lockhart Clarke, which is there given (p. 47).

In examining the fresh brain, small portions may be removed on the end of a knife, placed upon the glass slide, and moistened with a little serum, or weak solution of sugar, but it must be admitted little can be learnt by such a mode of examination, as the relation of the structures to each other is completely destroyed. For examining the arrangement and distribution of nerve fibres, portions of brain should be hardened in the chromic acid solution, when very thin sections can be obtained with a sharp razor. Dilute solution of caustic soda is also exceedingly useful for rendering the nerve fibres more distinct. The minute anatomy of the brain may be studied in man and in the higher animals in specimens prepared by injection and staining.

I have obtained beautiful preparations of brain as follows :—A large artery of the pia mater is selected, and when a fine injecting pipe has been introduced, carefully tied to the pipe. That vessel is injected with strong carmine fluid. After twenty-four hours, a little glycerine and water is thrown in, and this is followed by blue injection. Small pieces are then to be removed and placed in glycerine, to which a little chromic acid and bichromate of potash glycerine (page 103) have been added. After soaking for two or three weeks, thin sections may be made with a very sharp knife and transferred to glycerine containing acetic acid (10 to 15 drops of glacial acetic acid to the ounce). The sections are to be gently pressed, removed from the slide, and transferred to another drop of acetic acid glycerine. This process is to be repeated until the section is sufficiently frayed out to enable the observer to see the delicate nerve fibres crossing one another, and passing in every direction. When a good specimen is obtained, it may be placed under the $\frac{1}{25}$ or $\frac{1}{50}$ of an inch object-glass, and the continuation of the fibres with the tissue of the cells will be distinctly seen. The latter may be traced for a long distance from the cells, dividing and subdividing as they go. Many of the finest fibres are much less than $\frac{1}{100,000}$ of an inch in diameter, but in fortunate specimens they may be traced for a considerable distance.

In order to follow the course of the fine fibres for a long distance from the cells, I have adopted the following expedient. Clean sections of a convolution in various directions are made and soaked for a few hours in a solution of acetic acid and equal parts of glycerine and water (50 drops of acid to an ounce of the diluted glycerine). The sections are then transferred to plain glycerine and water, and after having been well washed in this fluid, are placed in the carmine fluid for from twelve to twenty-four hours. They are then again washed and afterwards removed to strong glycerine, with five drops of acetic acid to the ounce, and a little of the chromic acid and glycerine. When they have soaked

for a week or two, very thin sections are to be removed and examined in the usual manner. If the operations have been successfully performed, the finest fibres will be found to have been well stained of a bright red colour. Many may be followed for long distances, and the fibres resulting from division and subdivision will be seen crossing and interlacing with fibres taking different directions. In many cases it will be observed that after passing a certain distance as an apparently single thread, an excessively fine nerve fibre divides into two still finer fibres, which take opposite directions. This same remarkable fact is observed in the arrangement of nerve fibres in every part of the body. Whether we examine nerve trunks, nerves near centres, or in peripheral parts, or the ramifications of nerve fibres in the brain, spinal cord, or ganglia, we are struck by the fact that the fibres resulting from the splitting of a trunk, or the division of what seems to be a fine fibre, invariably pursue opposite directions.

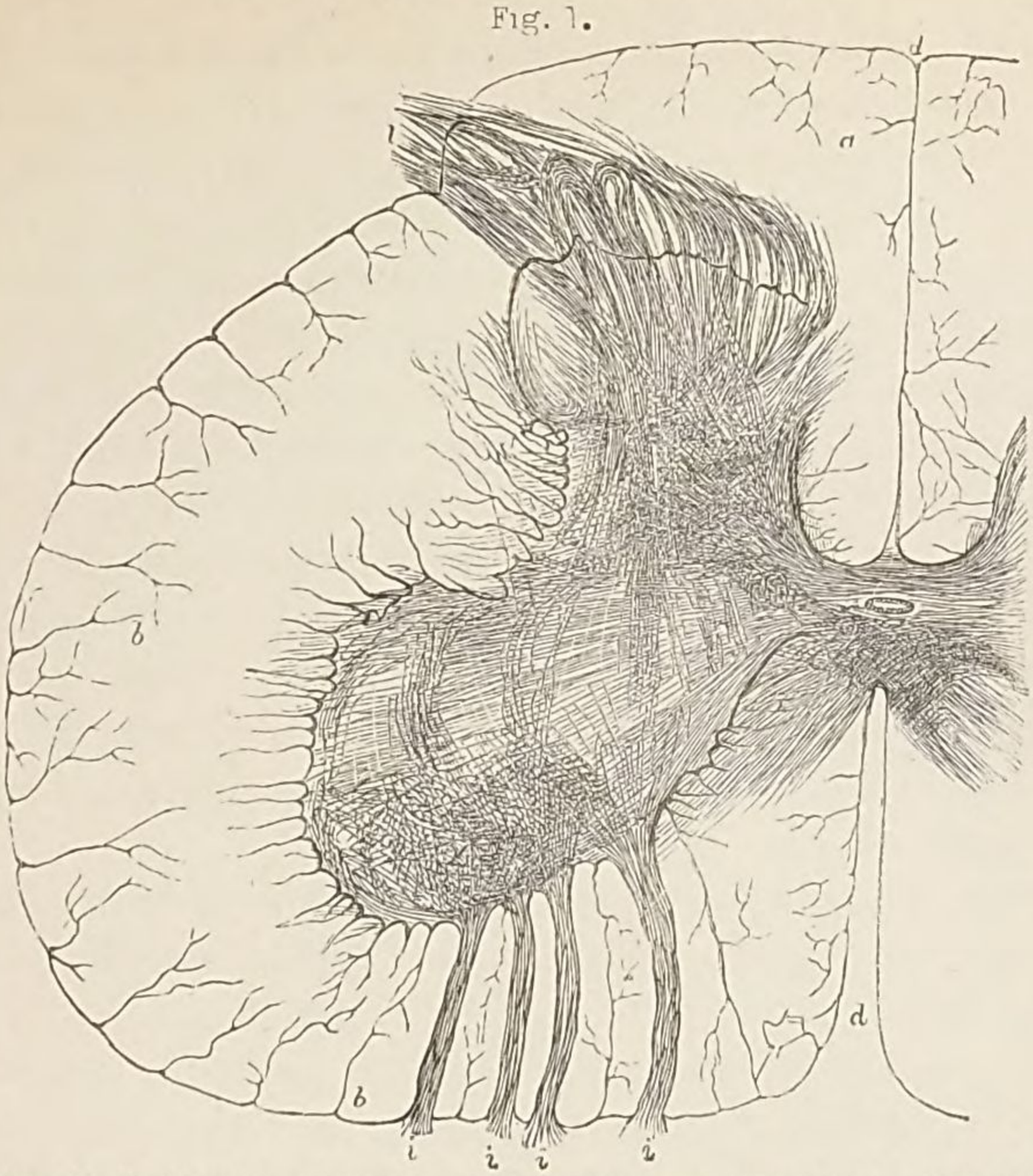
If a portion of white cerebral matter be treated with water, the nerve fibres soon become changed in character, apparently in consequence of the partial separation of the oily from the albuminous constituents of which the myelin or white substances of Schwann consists. The oily matter forms distinct and separate globules, often of considerable size or it tends to collect in quantity in different parts of the fibre, producing a beaded appearance. A similar change takes place in nerve fibres generally, if they are not examined very recently, or if they have been soaked for a short time in water. In figs. 3, 4, 5, 6, pl. XXI, p. 190, some of these changes are represented.

393. Examination of the Spinal Cord.—Different parts of the cord may be examined in the fresh state, but in order to demonstrate the beautiful structure described and figured in modern works, we must have recourse to the methods of preparation already described. Segments of different parts are to be placed in the solutions, and allowed to harden, when very thin sections may be readily obtained and examined, fig. 1, pl. LXVIII. These are to be preserved according to the plan many times referred to.

Specimens of the spinal cord may be prepared according to the plan given in page 425, for preparing very thin sections of the brain. These same methods of preparation are also applicable to the investigation of the minute structure of the ganglia and nerve plexuses in various parts of the body. Preparations of the brain and spinal cord in various cases of disease are to be made in precisely the same manner as has been recommended above. Specimens of the nerve textures in disease may be preserved in glycerine quite as successfully as those of the healthy tissues.

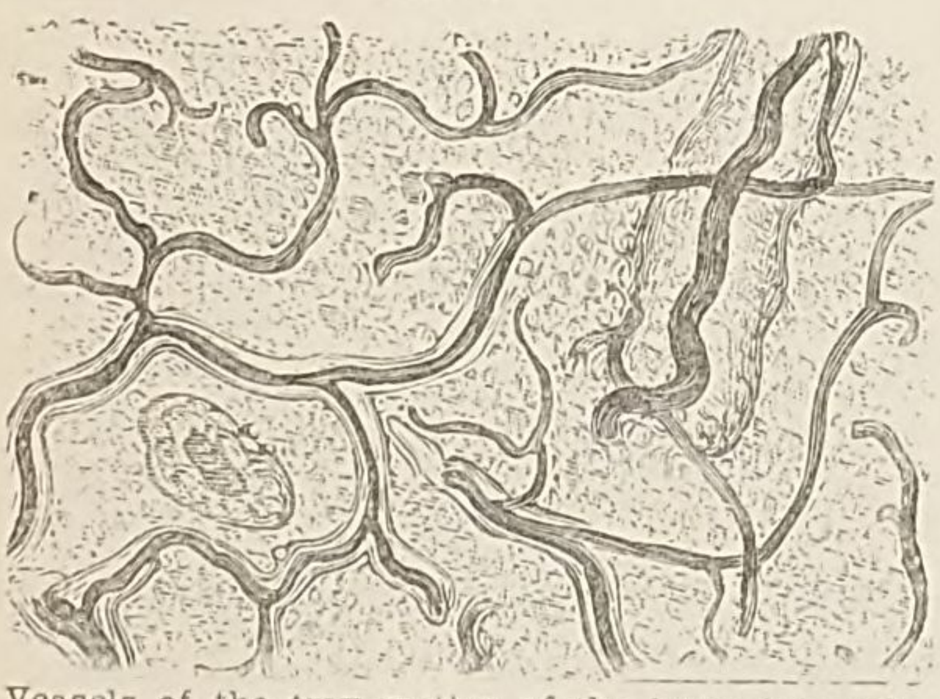
394.—On Ascertaining the Specific Gravity of the Brain.—As much attention has been paid to the density of the brain in various cases,

Fig. 1.



Transverse section of half of lumbar enlargement of cord, showing course of fibres. After LOCKHART CLARKE. p. 426.

Fig 2.

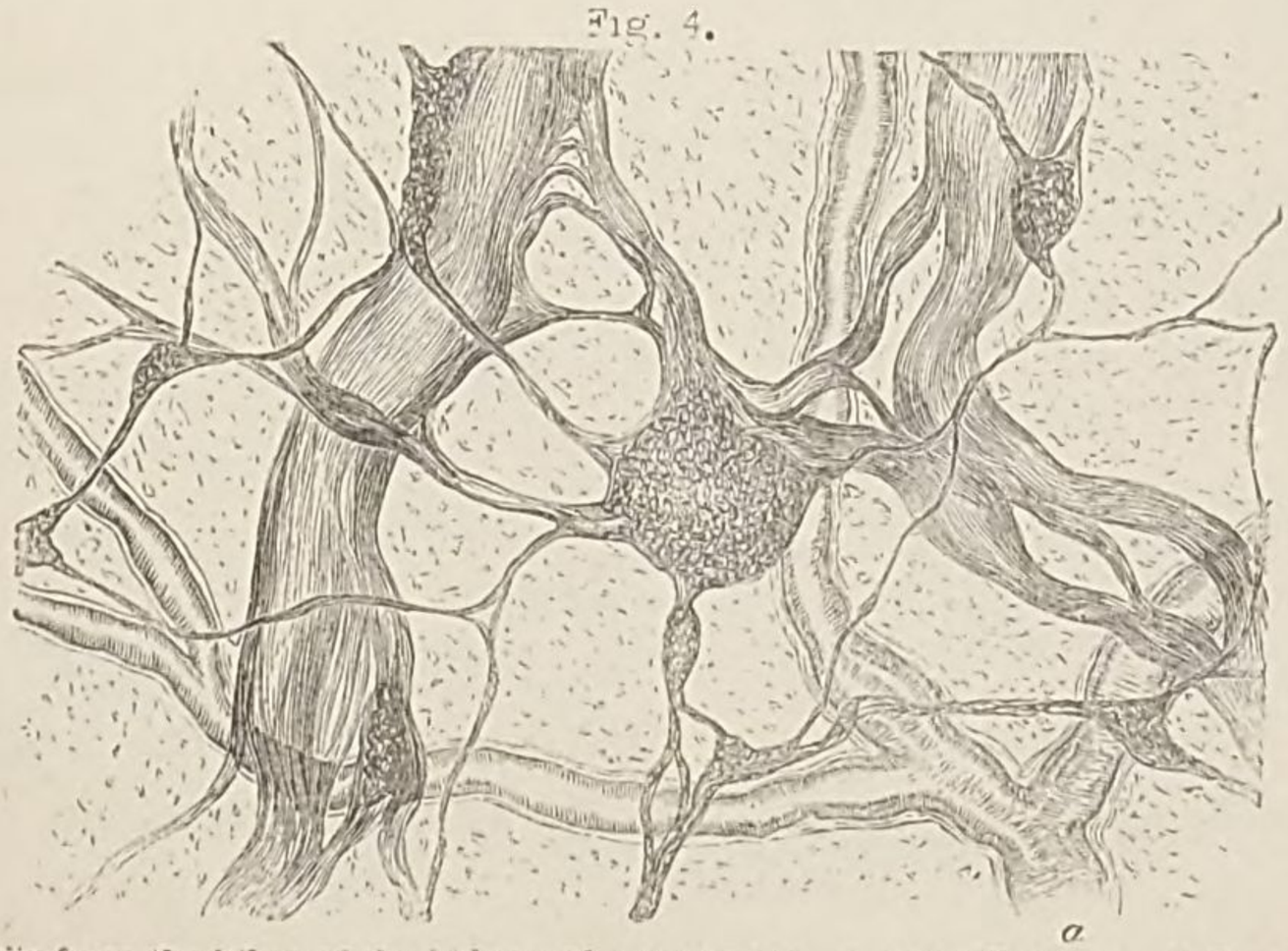


Vessels of the grey matter of the brain injected Specimen mounted in balsam. × 215. p 424.

Fig. 3.

Vessels of grey matter injected. Preparation mounted in glycerine. × 215. p. 424.

Fig. 4.



Ganglia from the hilus of the kidney of a young pig. *a*, a small artery × 30. p. 429.

1000th of an inch |___| × 215.

[To face page 426.

and as these investigations if followed up are likely to lead to very important conclusions, I think it desirable to describe here the method of pursuing such enquiries.

The researches of Dr. Bucknill, "Lancet," December 24, 1852, Dr. Sankey, "British and Foreign Med. Chir. Review," January, 1853, and others, have shown that the density of the brain varies considerably in different conditions. The specific gravity of the entire organ is in many cases affected, but it is obviously of the first importance to ascertain the density of the different portions separately. In this manner various parts may be proved to have suffered in nutrition, although no structural changes can be detected, even with the microscope. The specific gravity of the entire brain in health is about 1039; in cases of paralysis it is much higher, but varies from 1036 to 1050.

From very numerous observations Dr. Sankey has ascertained that the average specific gravity of the grey matter is 1034 in both sexes, while the mean specific gravity of the white matter is 1041.

Dr. Aitken, "Glasgow Medical Journal," No. I, 1853, has ascertained the specific gravity of the central parts of the brain to be as follows: the central ganglia 1040 to 1047; the cerebrum from 1030 to 1048; the cerebellum from 1038 to 1049. In a case of chronic hemiplegia the specific gravity of the corpus striatum and optic thalamus on the right, or sound side, was 1025, while the same parts on the left or paralysed side were 1031. It is desirable that the specific gravity of the different parts of the brain in health should be ascertained by an extended series of observations, as it is probable that very important results would be arrived at by comparing the numbers with those obtained in cases of disease.

The percentage of solid matter in different parts of a brain which may be concluded to be healthy, is shown in the following note.*

* The brain was obtained from the body of a man who was in good health at the time, and was killed by falling from the top of a house. He died about eight hours after the fall.

White matter of cerebellum—	
Water	67·27
Solid matter	32·73
White matter of hemispheres—	
Water	69·45
Solid matter	30·55
Medulla oblongata—	
Water	73·75
Solid matter	26·25
Optic thalamus—	
Water	74·60
Solid matter	25·40
Grey matter, cerebellum—	
Water	79·94
Solid matter	20·06
Corpus striatum—	
Water	79·96
Solid matter	20·04
Grey matter of convolutions—	
Water	80·58
Solid matter	19·42

The following cases show how the proportion of water and solid matter may vary in disease:

1. Brain of a child aged six weeks. Cause of death unknown. Body generally

Method of ascertaining the Specific Gravity of the Brain.—The specific gravity is ascertained by placing little pieces of the brain, about the size of a small nut, in solutions, the density of which has been previously taken. A number of saline solutions are prepared, varying in specific gravity from 1025 to 1055.

Solutions of chloride of sodium were first employed, but Dr. Aitken recommends sulphate of magnesia. Glycerine would also answer the purpose well, but the expense of the solution would be too great. The same portion of fluid should not be used for more than one or two experiments.

The salt is dissolved in a considerable quantity of water, and the density of the solution ascertained with an accurately graduated hydrometer, or with the specific gravity bottle. To a portion of this solution more water or salt is added, as the case may be, and the specific gravity is again ascertained as before. Portions of this are diluted until we have prepared a number of solutions, which may be preserved in separate bottles, each having the specific gravity of the solution it contains marked upon it. When an experiment is made, a little of each solution is poured into small glasses placed in regular order; the piece of brain is to be placed in one, and if it rises to the surface it must be tried in the next lighter one above it; but if it sinks to the bottom it must be removed with forceps and placed in a more dense solution. After a few trials, a solution will be found in which the morsel neither sinks to

well nourished; viscera all healthy; brain very soft, though examined within eight hours after death—of a waxy appearance. 2. Brain of a girl æt. nineteen, who died of diabetes. The brain was very firm, and no morbid appearances were observed. The white and grey matter contained—

Water	89·60	Water	74·85
Solid matter	10·40	Solid matter	25·15

3. Brain of a woman aged forty, who died of apoplexy. White matter of cerebrum apparently healthy. 4. Softened cerebral matter surrounding the clot.

Water	71·4	Water	81·49
Solid matter	28·6	Solid matter	18·51

5. Brain of a girl aged about twelve, who died from induration of a portion of white matter of the anterior lobe of the left hemisphere, about the size of a walnut. 6. White cerebral matter from the opposite side, and from the same side as that in which the indurated portion was situated.

Indurated portion—			
Water	75·24	Water	80·29
Solid matter	24·76	Solid matter	19·71

7. Brain in which there was an indurated portion in the anterior part of one hemisphere. 8. Anterior portion of opposite hemisphere, which was healthy. Specific gravity, 1044.

Indurated portions sp. gr. 1042—			
Water	81·59	Water	70·29
Solid matter	18·41	Solid matter	29·71

—"Archives of Medicine," No. II, page 155.

the bottom, nor swims on the surface. The weight of equal bulks of the brain and of the fluid is the same, and the specific gravity of the fluid used, which is known, indicates that of the brain, which is required.

395. Sympathetic Nerves and Ganglia.—For the study of the arrangement of the nerve fibres of the sympathetic system I recommend the frog, the white mouse, and certain tissues of the human subject. In the areolar tissue between the mucous and muscular coats of the intestinal canal, multitudes of ganglia with many entering and issuing nerve fibres will be found. In the mouse these are very delicate and transparent, but of large size, and can be very clearly demonstrated. In the corresponding situation, in the human subject, the ganglia will be demonstrated without difficulty. Many of them are large, and contain as many as twenty cells; some are very small, and consist of three or four only. Intercommunicating trunks of nerve fibres exist in considerable number, and bundles of fine nerve fibres may be traced to their distribution in the mucous membrane and the villi, and others to the muscular coat. In pls. LXVIII, IX, will be found some drawings of ganglia and the nerve fibres entering and leaving them at different points, and are well seen. The number of such nerve centres and intercommunicating nerve cords in connection with the important organs in the chest and abdomen is remarkable, and anyone who can picture to himself the true arrangement, say in connection with a square inch of intestine, will feel astonished that an apparatus of such marvellous extent and delicacy sustains the constant shocks and disturbances to which it is exposed, and works on as it does in many instances for nearly a century with scarcely a break. Not only so, but it withstands great shocks and disturbances in disease, suffering permanent damage only from the very long continuance of morbid conditions.

In a case of death from capillary hæmorrhage from a very large extent of small intestine consequent upon cirrhosis of the liver, I found the ganglia and nerve fibres much altered. The substance of the ganglion cells was granular, and numerous oil globules were interspersed through them, and a similar change had affected the nerve fibres. These ganglia are probably concerned in governing the calibre of the little arteries distributed to the several tissues of the intestine, and also in regulating the degree of contraction of the muscular fibres of the muscular coat under varying circumstances.

Skin and its Appendages.

396. Cuticle.—The cuticle may be subjected to examination either by scraping the surface, in which case only the most superficial cells, which are often not well defined, will be obtained; or by making a thin section of dried or hardened cuticle, with a sharp knife. If a portion of skin be allowed to remain for some days in a moist atmosphere, the

cuticle will become separable in large flakes, fragments of which may be moistened with water or glycerine in the usual way, and subjected to examination. The epithelial lining of the sweat glands will often be drawn out from the tubes adherent to the deep surface of the cuticle.

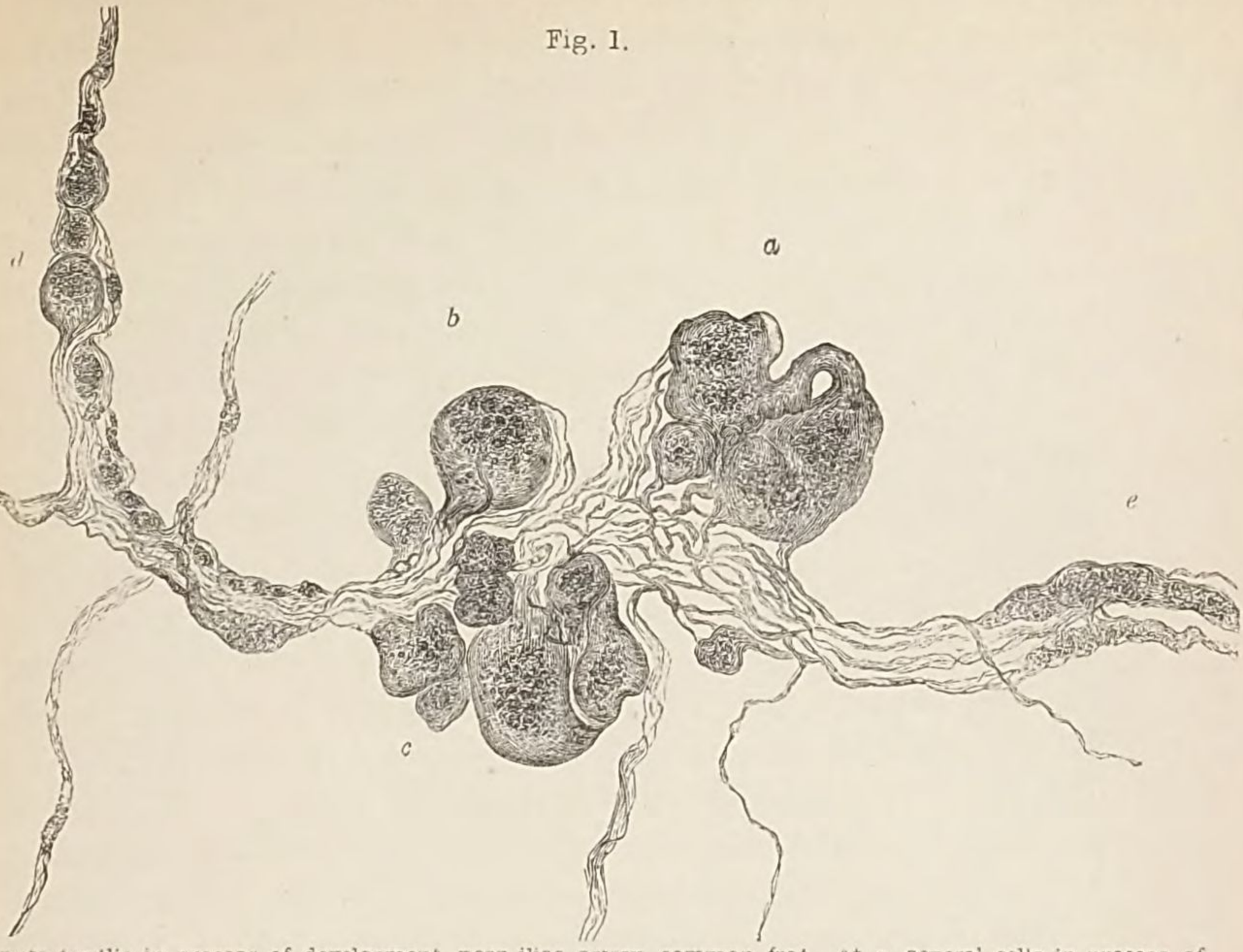
The epithelium is seen to differ in character according as it is taken from the deep or from the most superficial layers of cuticle. In the former situation the epidermic cells are more or less rounded in form, while on the surface they are flattened and adhere to each other, forming small scales, in which the original form of the cell is with difficulty made out. The deepest layer of the cuticle consists chiefly of a few small cells, with round or oval bioplasts, which gradually become converted into cells. It is in the cells in this situation that the colouring matter is deposited in the dark races, and it was to this portion of the cuticle that the term *rete mucosum* was applied. As the cells approach the surface, the colouring matter appears to diminish in quantity, owing probably to changes taking place in the chemical nature of the material. The bioplasts in the deeper layers of the cuticle are rendered transparent by and are nearly soluble in acetic acid, while those on the surface are unaffected by this reagent.

Upon examining the under surface of the cuticle, which has been removed as above directed, it will be found to present several depressions, in which the tactile papillæ of the cutis are lodged. Upon removing the cuticle by maceration, from some situations, such as the palm of the hand, or heel, or from the anterior surface of the leg, the epithelial lining of the sweat ducts as they pass through the cutis, will often be found adhering firmly to it. Preparations of this kind may be preserved in glycerine, to which a very little acetic acid has been added.

As is well known, after scarlatina, the superficial layers of the cuticle separate in the form of scales, which are sometimes of large size. The old cuticle of the sole of the foot and the palm of the hand is sometimes detached without a break. Amongst the cuticular cells upon the deep aspect may sometimes be found collections of minute bioplasts, which are probably the contagious particles which are the agents concerned in propagating the disease, pl. LXX, fig. 2.

Pigment Cells.—The cells containing pigment are very readily demonstrated in the skin of the negro, in that of several of the lower animals, or in the freckles which may often be obtained from different parts of the body of some subjects. A preparation of the cuticle of the negro may be preserved in Canada balsam. The cuticle may be separated as above described, dried flat between folds of clean paper, or between two plates of glass, and mounted in the usual way. The method of preparing a vertical section of the cuticle is described in page 432.

Fig. 1.



Minute ganglia in process of development, near iliac artery, common frog. At *a*, several cells in process of division. At *c*, such cells at a still earlier period. At *d*, several cells and fibres are being formed. *e*, masses of bioplasm in the course of nerve fibres becoming developed into ganglion cells. × 700. p. 429.

Fig. 2.

Ganglion cell and nerve fibres, sympathetic Ox. × 215 p 429.

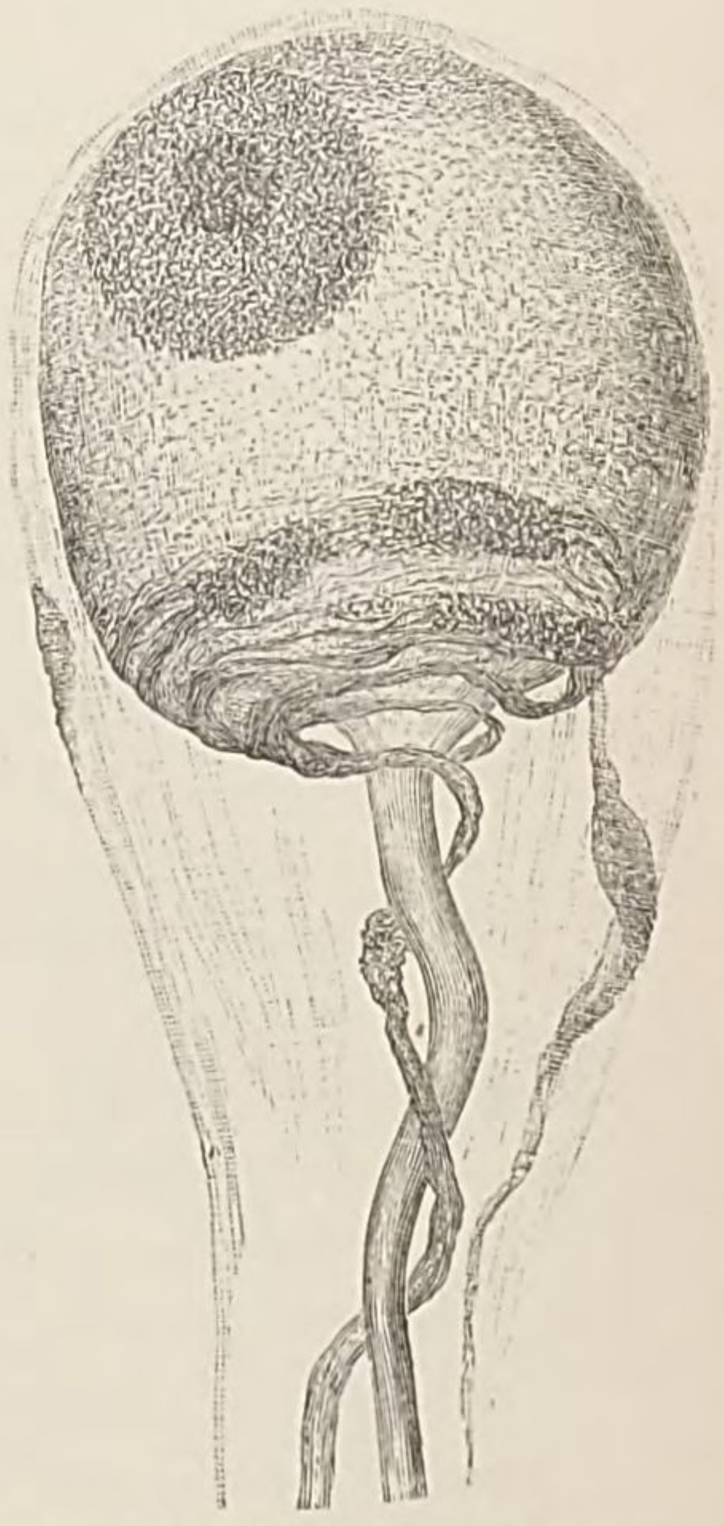
Fig. 3.

A small portion of ganglion of the sympathetic of a boy. × 700. p 429.

Fig 4.

Young ganglion cell from the sympathetic of the hyla. × 700.

Fig. 5.



Fully formed ganglion cell from the hyla. × 700. p. 429.

1000th of an inch ⌊_____⌋ × 700.

[To face page 430

The walls of the vessels of the peritoneum, lungs, &c., and the skin, of the frog, contain beautiful varieties of very dark pigment cells, which consist of several branches of irregular form radiating from the central part of the cell. These anastomose with the ramifications of neighbouring cells.

397. Papillæ.—The papillæ may be shown in two ways, either by making a vertical section of the skin previous to the removal of the cuticle ; or the true skin only may be hardened and a section made.

The best situations from which to take the skin for the purpose of examining the papillæ, are the palmar surface of the hand and fingers, and the sole of the foot. After the skin has been soaked for a time in water, and the cuticle removed, a transverse section, including the papillæ, may be made as follows : a gentle stream of water is to be allowed to flow over the papillæ, in order to make them all fall in one direction. This is readily effected by inclining the piece of skin downwards while the water is running. After being drained, a cut is made with a very sharp knife across the piece of skin in its upper part, in a direction at right angles to that which the papillæ have been caused to assume. Upon now turning the preparation so that the freshly-cut surface is in the most dependent position, and allowing the jet of water to flow, the direction of the papillæ will be reversed, and it is obvious that a very thin section off the freshly-cut surface will include one or more rows of *entire* papillæ. The section may then be examined, and may be preserved in liquid ; or it may be placed upon a glass slide, gently dried, and mounted in Canada balsam. The papillæ of the skin of the foot of the dog are large and well-marked.

The beautiful specimen of papillæ represented in figs. 3, 4, plate LXX, from the teat of a cow suffering from cattle plague, was prepared according to the principles laid down in Chapter VII, the staining being effected by injecting the vessels with carmine fluid, and the Prussian blue being injected as soon as the bioplasm had been properly coloured. After small pieces had been soaked for two or three weeks in glycerine the cuticle was removed without difficulty, and thin sections were easily obtained which could be examined with the highest powers.

In order to examine the structure of the papillæ at once, a tolerably fresh specimen of skin should be chosen, and as thin a section as possible should be made. The specimen may then be treated with weak caustic soda, or with a little acetic acid, and subjected to examination. It is in this way that the nerves may occasionally be demonstrated in the papillæ, and frequently the vascular loop may be thus rendered distinct ; but the arrangement of the vessels is always better shown in a specimen injected with an opaque injection.

The "axis-corpuscles," or touch-bodies may be shown in the papillæ situated at the tips of the fingers, or in the palm of the hand, by treating

a thin section specimen with strong acetic acid. A papilla may be met with in this situation terminating in two or three points. One of these will perhaps contain a touch-corpuscle, while in the others only a vascular loop can be seen, and no nerve fibre whatever can be distinguished. These so-called touch-bodies invariably have more than one nerve fibre connected with them, and the "body" itself consists, I believe, of numerous convolutions of fine nerve fibres, much bent and compressed so as to form an oval mass—multitudes of oval bioplasts connected with the nerve fibres are seen running across the corpuscle. I have obtained good specimens from sections of the skin of the finger of a monkey which had been soaking for some time in glycerine.

I have prepared beautiful specimens of the papillæ containing touch-bodies from the skin of the tip of the finger of a child, which had been injected first with the carmine fluid and afterwards with Prussian blue fluid (page 105). In preparations thus made it can be seen that the nerve fibres are folded transversely throughout the corpuscle, long oval bioplasts or nuclei being connected with the nerves and lying transversely across the corpuscle.

398. Method of making a Verticle Section of Skin.—In a section of this kind all the structures entering into the formation of skin can be demonstrated by the student very easily, and the arrangement of the hair-bulbs and sebaceous follicles may also be seen if the skin be taken from a part in which these structures abound. The disposition of the sweat ducts and arrangement of the glands may be well shown in such a preparation. It is exceedingly difficult to cut a section of skin in the recent state sufficiently thin for observation; hence, if the student has not mastered more elaborate methods of preparation, he may resort to the old plan which is carried out as follows: the skin should be perfectly fresh, and a piece about two inches square, or rather less, may be stretched, with the outer surface downwards, upon a thick deal board, by means of numerous pins. If the sudoriferous glands are to be included in the preparation, care must be taken to leave sufficient of the subcutaneous areolar tissue. The piece of skin is allowed to dry by exposure to the air. Several small pieces, taken from various parts of the body,—scalp, eyelids, chin, mamma, axilla, arm or leg, palm of the hand, tips of the fingers, scrotum, sole of the foot,—may be pinned out on the same board, care being taken to attach a label to each. In these, the varying thickness of the epidermis and true skin, and other peculiarities in the different regions may be demonstrated. The portion of skin, being quite dry, it is to be removed from the board, and, after cutting off the edge, several thin sections may be made, by the aid of a very sharp knife, through its whole thickness. In order to obtain a good specimen of the spiral portion of the sweat ducts, traversing the epidermis, the skin of the heel should be selected, and the section

should be made parallel with the furrows, and in a slightly slanting direction, instead of at a right angle with the surface.

If more detailed investigations are desired, particularly if the arrangement of the nerve fibres is to be followed out, pieces of skin should be prepared according to the recommendations given in Chapter VII. It is not difficult to find an artery large enough to receive a small pipe for injecting the vessels.

Potash or caustic soda much diluted are valuable re-agents in the microscopic examination of skin. Carbonate of potash has also been used. Sections of skin may often be preserved very well in a solution of chloride of calcium.

Skin may be macerated in dilute nitric acid (one of acid to two of water) for twenty-four hours, when the sweat glands are easily distinguished upon making a section (Giraldès, quoted by Kölliker).

The most satisfactory specimens for examining the intimate structure of the skin with high powers, are obtained from skin prepared according to the plan recommended in Chapter VII. After staining and injection, small pieces are placed in glycerine, with a few drops of chromic acid and bichromate of potash (p. 103). After some weeks the tissues will be found of the right degree of hardness to furnish sections.

399. Corns, and other growths, which consist of thickened epidermis, and warts in which the corium is much thickened and the papillæ enormously enlarged, may be subjected to examination, and sections obtained in the same way as directed for making sections of skin. A curious horny growth from the nose is figured in pl. LXXI.

In disease the subcutaneous areolar tissue is sometimes found thickened over a considerable extent, or over small circumscribed spaces; in which latter case the sensation of small subcutaneous tumours is produced if the affected portions of skin be pinched up between the finger and thumb. The condition termed *Elephantiasis* appears to consist of a thickening and exaggerated growth of the subcutaneous areolar tissue, and the pouring out into the areolæ or interspaces of lymph, which subsequently becomes organised and a part of the tissue. Sections of skin in this state may be made after being hardened in alcohol or in a solution of chromic acid.

400. Nails.—The structure of nails may be studied in sections cut in various directions with a very sharp knife. These may be soaked for some time in glycerine and then immersed in glycerine containing ten drops of acetic acid to the ounce. But the most instructive specimens are obtained by injecting one of the arteries of the finger with carmine fluid, and then making sections through the matrix and the nail tissue. In transverse sections the vessels will be seen beautifully injected, and the youngest bioplasts of the nail deeply stained with carmine on the upper surface.

In old age a remarkable change often takes place in the nail—a very redundant growth of nail tissue being formed. The nail soon loses its peculiar characters and assumes those of a claw. Growth takes place from the whole surface of the matrix, though faster at the root than at the free extremity, owing to which circumstance the nail is curved downwards like a claw. The tissue accumulates so that the nail becomes of great thickness and length. The altered nail may be two inches or more in length, firm, solid, and curved after the fashion of the over-grown claw of a cage bird. Some very interesting examples of this change are represented in pl. LXX, fig. 5, the original specimens having been given to me some years ago by a kind friend, but whose name I regret to say I cannot call to mind.

401. Sebaceous Glands.—The sebaceous glands are very easily demonstrated. Their arrangement is made out very distinctly in vertical sections of the skin of the scalp of the fœtus. I obtained some very beautiful specimens from skin which had been hardened for some time in acetic acid. The hairs and hair bulbs are also often well shown in the same specimen. The skin of an eight months' fœtus injected and stained with carmine affords beautiful specimens, showing very clearly the several structures of which the skin consists.

402. Sweat Glands.—The sweat glands are also demonstrated most readily in the skin of the fœtus. The gland itself may be easily dissected out from the subcutaneous areolar tissue of the axilla, in which locality the glands are of large size. They may be preserved in glycerine. The course of the ducts through the cuticle should be noticed, especially in the cuticle of the heel, where they pursue a spiral course. If the skin be removed from the heel and allowed to dry, sections can often be obtained in which the connection between the spiral portion of the duct in the cuticle and that part which passes through the true skin, may be demonstrated. These sections require moistening with water, and they may be mounted in a solution of chloride of calcium, or naphtha and creosote fluid. Glycerine renders them very transparent.

403. Hairs.—The structure and mode of growth of hair may be well observed in sections of the skin of the fœtus. The mode of obtaining transverse and longitudinal sections of hair has been described in "How to Work with the Microscope." The moist structures surrounding the bulb of the hair (root sheaths) when drawn from its follicles, are well seen in specimens which have been soaked in glycerine, and the cells in the medulla of hair are also best displayed in the same manner. The fibrous structure of the hair is sometimes well seen at the ends where the fibres are separated from each other, and not unfrequently, in hairs which have been twisted for some time into knots, the hard elongated fibres of which they are composed have

Fig. 1.

Ganglia and nerve trunks near iliac artery. Common frog. Many undergoing development. × 40. p. 429.

Fig. 2.

Cells of cuticle from the teat of a cow suffering from cattle plague, showing the morbid bioplasm in and amongst the epithelial cells × 212. p. 430.

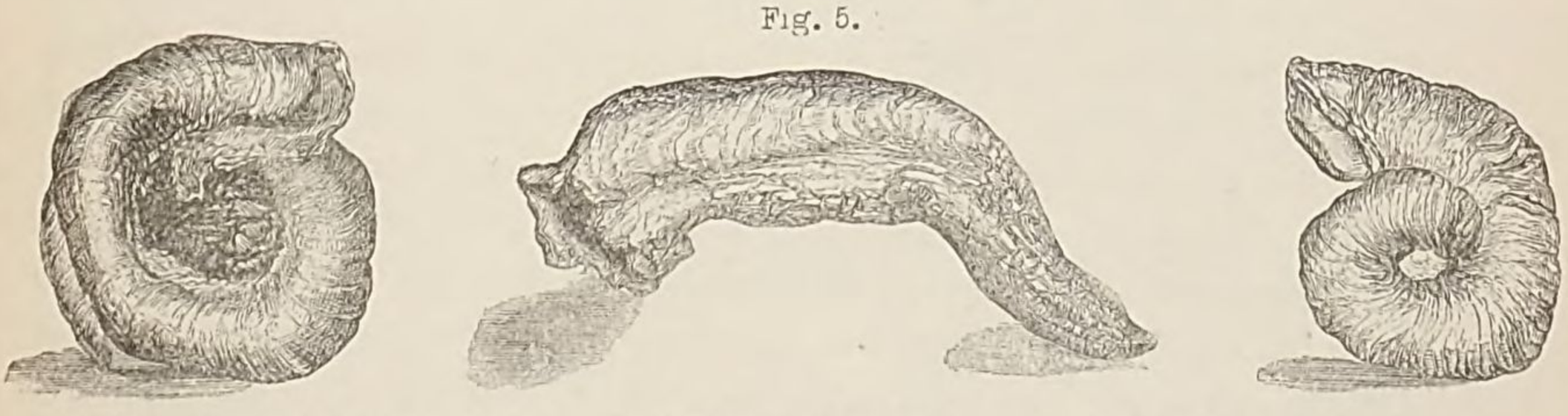
Fig 3.

Section through cuticle and cutis of the teat of a cow, showing vessels, connective tissue, and epithelium. × 130. p. 431.

Fig. 4.

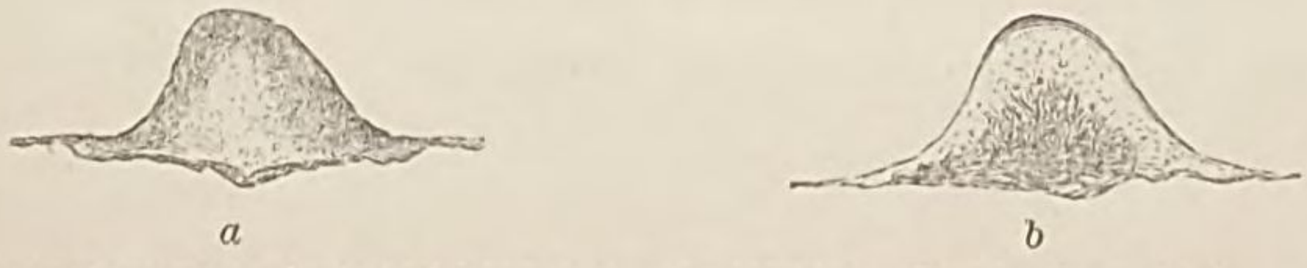
Papillæ as in Fig. 3, from which the epithelium has been removed. × 130.

Fig. 5.



Three nails from the toes. Taken from an old woman in a workhouse. Of the case, unfortunately, I have no note. Natural size. p. 434.

Fig. 6.



Horny growth from the nose, which had been one year growing. Removed by Dr. Osborne, of Bitterne, 1869. *a*. surface. *b*. section of the tumour. Natural size. p. 433.

[To face page 434.

become unravelled as it were, and the manner in which these are combined so as to form the shaft of the hair is well displayed. A drawing of a diseased hair which is much split up, is represented in pl. LXXXII, fig. 3.

404. Molluscum.—Upon carefully examining the tumours from a case of molluscum in different stages of growth, I was led to the conclusion that they really consist of altered structures connected with the hair bulb. The specimens I prepared were certainly not due to a morbid state of the sebaceous glands as used to be supposed. My drawings representing the altered hair-follicles accompany a paper upon the subject published in the Reports of the Pathological Society for 1855, page 313. The conclusions I arrived at were as follows:

1. That neither the sebaceous glands nor the sweat glands, nor their ducts, were concerned in the formation of the tumours.

2. That the tumour consisted essentially of a morbid alteration of the structures concerned in the formation of the hair, especially of the cells at the deepest part of the follicle, and of the follicle itself.

3. That the subcutaneous areolar tissue was considerably hypertrophied; both its white and yellow elements being coarser and more abundant than in health. "Trans. Path. Soc., 1855," p. 313.

Many tumours connected with the skin depend upon an altered growth of the hair. I have seen hairs coiled up in the centre of small fibrous tumours beneath the skin. The follicle seems to have been closed at its summit, and the egress of the hair being prevented, it gradually becomes twisted up in consequence of its growth still continuing from the root. The areolar tissue and other structures in the neighbourhood become gradually involved in the alteration, and the tumour as it increases in size becomes more complicated in structure, so that its real nature is not very easily made out. No one who considers the many different anatomical structures entering into the formation of the hair sac, and in its neighbourhood will feel surprised at this. The different character of these tumours is readily explained by the circumstance that in one tumour the growth of one structure, in another the growth of a different one preponderates over the growth of the rest. Such morbid growths are to be prepared for examination according to the same principles laid down for the examination of the healthy skin and its appendages.

405. Nose.—Much yet remains to be discovered concerning the minute structures of the olfactory portion of the mucous membrane of the nose. It rapidly undergoes post mortem change, and it is, therefore, necessary to examine it as soon after death as possible. The upper brown portion, near the ethmoid bone, is the part of the membrane which should be especially examined, for it is there that the fine ramifications of the olfactory nerves will be found, and this is the region in which odorous particles come into contact with the peripheral organ

and bring about those physical changes which enable us to distinguish one smell from another. The student should obtain specimens of the mucous membrane from the sheep, as the head of this animal can always be obtained perfectly fresh. It is to be sawn through longitudinally, a little on one side of the centre, and the delicate mucous membrane carefully removed from the laminæ of bone which it covers. With the aid of a very sharp knife, or scissors, thin pieces may be obtained fit for examination. Care must be taken to prevent much pressure by the thin glass cover. The epithelium of the sinuses and lower part of the nose is columnar and ciliated; that near the orifice is scaly, and approaches in character to that of skin; while in the *olfactory region* it is not ciliated, but consists of a thick layer of small granular cells (Todd and Bowman's Physiology, vol. ii. p. 5). The nerves are destitute of the white substance of Schwann, and take the form of delicate flattened bands, with oval nuclei scattered at tolerably equal intervals along them. The termination of the nerves has not been ascertained, although there can be no doubt that delicate nerve filaments may be followed up to the epithelium-like cells. Between the latter very fine threads may be discerned, and, according to the recent observations of Eckhard and Ecker, may be traced beyond the surface of the epithelium. In other instances, the nerve filaments appear to terminate in epithelial cells, or in bodies not to be easily distinguished from these. The mucous membrane is highly vascular, and the capillaries form many dilated loops which were first noticed by Quekett. The structure of the minute anatomical elements in the olfactory region of the nose is worthy of very careful and extended investigation.

406. Ear.—The investigation of the minute anatomy of the ear is exceedingly difficult. The nervous structure is so delicate, that it undergoes very rapid changes after death. In investigations upon any special point, in the anatomy, it is better to examine the ears of animals, and afterwards compare the results with those obtained from researches on the human ear. Mr. Toynbee gives the following directions for removing the internal ear from the human subject. These will be found useful to the student of this branch of morbid anatomy.

"The simplest method of removing ears for the sake of dissection is, in the first place, to saw off the calvarium in the usual way, and then to take out both the petrous bones together, by means of two transverse vertical sections, one in front of each petrous bone, and the other posterior to it. The anterior of these sections should pass in a line a little anterior to the anterior clinoid processes, and the posterior in a line through the posterior third of each mastoid process. By means of these two sections, the trumpet-shaped extremity of each Eustachian tube, a portion of the mucous membrane of the fauces, and the whole of each petrous bone, together with the mastoid process, can be taken out. The

disadvantage of this procedure is the disfigurement which is apt to ensue from the falling in of the face. To avoid this disadvantage, another mode of removing the ears may be resorted to : this consists in taking out each petrous bone separately in the following manner :— The calvarium having been sawn off, an anterior section is to be made on each side on the same line as in the above plan, but extending only as far as the outer part of the body of the sphenoid bone ; a posterior section on each side is then to be made, as in the first plan, but not extending further inwards than the basilar process of the occipital bone. These two sections are to be made with a saw, or with a chisel and hammer ; the apex of each petrous bone is then to be separated from the sphenoid and occipital bones, and each petrous bone, the outer ear and integument being detached and reflected downwards, is to be drawn outwards, taking care, by inserting the scalpel deeply, to remove as much of the soft parts as possible. With this second plan there is a difficulty in removing the whole of the guttural portion of the Eustachian tube : with care, however, this portion may be removed, especially if the final sections separating the petrous bone from the occipital and sphenoid, be made to pass obliquely from above, downwards and inwards. The organ of hearing having been removed, the dissection may be conducted in the following manner :—The auditory nerve in the meatus should be first carefully examined, premising that a previous inspection has been made of the portion of the brain to which the *portio mollis* and *portio dura* nerves are attached. The size of the external meatus having been ascertained by allowing a strong light to fall into it, its anterior wall is to be removed by the cutting forceps, made by Messrs. Ash, of Broad Street, Golden Square ; the state of the epidermis, the ceruminous glands and secretion, the dermis, the periosteum, and bone, is to be noticed. The outer surface of the membrana tympani is then to be examined ; also the state of its epidermoid and dermoid laminæ, its degree of tension, and the amount of motion possessed by the malleus when pressed upon by a fine point. The next step is to ascertain the condition of the guttural portion of the Eustachian tube, to lay open the cartilaginous tube with the scissors, and then expose the cavity of the osseous portion by means of the cutting forceps. In doing this, the *tensor tympani* muscle is exposed ; its structure should be examined, and, if it has not a healthy appearance, portions of it should be submitted to microscopic inspection. The upper wall of the tympanum is next to be cut away by means of the cutting forceps ; in doing this, great care must be taken not to disturb or disconnect the malleus and incus, which lie immediately beneath it. After the tympanic cavity has been exposed, the first step is to draw the tensor tympani muscle, and to ascertain how far it causes a movement of the membrana tympani and ossicles. The incus and stapes are now to be touched

with a fine point, so as to ascertain their degree of mobility; the tendon of the stapedius muscle is also to be pressed upon. The condition of the mucous membrane of the tympanum, and of the mastoid cells, is then to be ascertained, and any peculiarity of the cavity, the existence of bands of adhesion, &c., to be noted. The most delicate part of the dissection, viz., that of the internal ear, must now be undertaken. The cavities of the vestibule and cochlea, are to be exposed by removing a small portion of the upper wall of each. Before reaching the vestibule, the superior semicircular canal will be cut through and removed; the membranous canal should be drawn out and inspected. As the cavities of the vestibule and cochlea are laid bare, it is desirable to see that the quantity of perilymph is natural, as well as its colour and consistence. The outer surface of the membranous labyrinth having been observed, it should be opened so as to expose the endolymph and otoconia, portions of all which parts should be removed for microscopic inspection. This having been effected, the remaining membranous semicircular canals are to be exposed, and the connection of the base of the stapes to the fenestra ovalis carefully examined. The last stage of the dissection consists in removing parts of the lamina spiralis, in examining them microscopically, and in exposing from within, by following the course of the scala tympani, the membrane of the fenestra rotunda. The only organ which now remains unexamined, is the stapedius muscle. In order to expose it, the course of the aquæductus Fallopii, beginning at the stylo-mastoid foramen, should be followed until the base of the pyramidal eminence, containing the muscle, is reached."—"A Descriptive Catalogue of Preparations illustrative of Diseases of the Ear," in the museum of Joseph Toynbee, F.R.S. Churchill, 1857.

407. Mode of preparing the Cochlea for Microscopical Examination.—My friend Dr. Pritchard sends me the following directions, which will be found very valuable:—

"The lamina spiralis may be examined in the fresh state, and for that purpose the cochlea of a new-born kitten is the most suitable, on account of the facility of getting at the lamina without injuring it; but these preparations are not of much service for showing the rods, and of course no vertical sections can be obtained from such soft and delicate tissues in the fresh state.

"The first point, and this is all-important, with regard to the preparation of the cochlea is that it should be taken as fresh as possible; immediately after death is of course the best, but within twenty-four hours will suffice if the brain has been removed early. The cochlea should then be taken from the animal, the stapes torn away, and the fenestra rotunda freely opened up. When the cochlea is deeply imbedded in bone, as is the case in the human subject and all the larger animals, as much as possible of the bone should be removed by the

saw, forceps, &c. The next object is to harden the membrane, and this may be accomplished by maceration in a solution of chromic acid about $\frac{1}{4}$ per cent. The cochlea may be kept in this for many months or only a few days, but as a rule three weeks to a month is the most satisfactory.

"In order to soften the bone, nitric acid should be used, from $\frac{1}{2}$ to 1 per cent., and this is best added to the chromic acid solution during the last few days of maceration. But the length of time required for softening the bone must of course vary with the thickness, &c., of the same; for the new-born kitten less than twenty-four hours will suffice; for the cat or dog two or three days; for the human, a fortnight or three weeks. It is very important to use a large quantity of the solution, or to change it every four or five days, whenever a cochlea of any of the larger animals is to be softened.

"When the bone is quite soft it must be prepared for section in the following manner:—Make a small conical bag of paper, fill it with strong gum-water, and put in the softened cochlea (taken directly from the chromic acid solution), leave it in the gum-water to soak for a few hours, and then place the whole (paper bag and all) in rectified spirit (methylated). At the end of twenty-four hours remove the bag from the spirit, and pick away the paper and gum, which will then be quite white and hard. Should the cochlea be now found not sufficiently firm, it may be steeped in absolute alcohol for a minute or two, and then it will be ready for section.

"Stirling's machine, with a very sharp razor, should be used for making the sections, and these must be floated in spirit.

"For holding the cochlea in the machine, Ferrier's mixture is the most convenient; it consists of

Parafine	5 parts.
Spermaceti	2 parts.
Lard	1 part.

Melted in a water-bath.

"Care must be taken in placing the cochlea in the machine, so that vertical or horizontal sections may be cut. Before mounting, the gum must be carefully dissolved away by water, and the preparations should be put up in glycerine, or, what is perhaps better, in a saturated solution of acetate of potash, made with camphor water."

408. Freezing Tissues prior to cutting thin Sections.—Dr. Pritchard has recently suggested a most convenient plan for freezing tissues and cutting very thin sections when congelation is complete. By this process thin sections of perfectly fresh tissues may be readily obtained. Dr. Pritchard's instrument consists of two parts—(1) a metallic cylinder fitted with a wooden handle; (2) a cap of thick felt.

(1) The metallic cylinder, which is solid throughout, should be made

of copper on account of its conductivity, but gun metal, or even brass, will answer the purpose sufficiently well. Its exact size or shape is not of very much consequence, so that it is large enough and convenient to handle. The following dimensions are recommended: diameter of metal cylinder 1⅜ inch, length 1¼ inch; the diameter of the end of the wooden handle should also be 1⅜ inch; the plug end should taper gradually, and the hole in the metal be a deep one, so that the plug may be pushed further in when the metal contracts on cooling. Both the wooden and metallic ends are made flat, so that the instrument can stand on either extremity; on the metal surface are a series of half a dozen concentric shallow grooves.

(2) Is simply a cap of thick felt, such as is used for boilers being preferable, made so as to fit somewhat loosely over the machine (1).

Mode of using the machine.—Plunge (1) with metallic face downwards into a mixture of finely pounded ice and salt; after remaining therein for three or four minutes, take it out and wipe with a clean cloth. The instrument has now been cooled down far below the freezing point, and on placing upon the metal plate a piece of soft tissue, this immediately freezes to the machine. The cap (2) must now be placed over the metal, but not allowed to touch the tissue, which will then freeze throughout in a very short space of time, varying according to the size of the tissue from a few seconds to one or two minutes. Now reverse the cap so as to leave the metallic top free, and, holding the whole in the left hand, cut the sections with the right by means of a sharp razor which has been kept cool in ice and water. Occasionally, the tissue may slip on the metal; when this is the case, remove the preparation, moisten it with gum-water, and replace it, when it will be found to adhere with much greater firmness. This slipping, however, very rarely occurs with perfectly fresh tissues, the grooves on the metallic surface tending to prevent it. The tissue will remain frozen quite long enough to make several score of sections, but should a thawing action set in, it may be covered with thin gutta-percha, and the machine again plunged into the ice and salt.

The advantages of this little machine are, first of all, its simplicity; and, secondly, the rapidity with which tissues may be frozen by its means. To illustrate the quickness of its action, it is only necessary to drop a little water on the cooled metal to convert it immediately into ice. Lastly, the apparatus may be made by any instrument maker or metal turner for a few shillings. The apparatus may be obtained of Mr. Baker, of Holborn, and Mr. Swift, of University Street, and other instrument makers. "Lancet," Dec. 11th, 1875. See also p. 442, in which another method of congelation is described.

Nerves of the Membrana Tympani.—For demonstrating the nerves of the membrana tympani, Dr. M. Watson recommends that the mem-

brane removed as soon as possible after death should be steeped for a few seconds in concentrated acetic acid. Then it should be placed in a solution of chloride of gold ·5 per cent., heated to about 100° for half an hour, and lastly soaked in glycerine, or in water slightly acidulated with acetic acid, and exposed to the light till it assumes a delicate purple colour.

409. Eye.—The microscopical examination of many of the tissues of the eye is a matter of great difficulty; and although it is impossible to describe at length the various plans adopted for demonstrating the anatomy of these different textures, it is nevertheless desirable to indicate a few of the operations which are employed in the investigation. The student should practise observations on the anatomy of the eye as much as possible; for if he is in the habit of investigating these structures he will be able to undertake any branch of microscopical inquiry with a fair prospect of success. The eye should be injected through a branch of the ophthalmic artery with the Prussian blue solution and in the course of a few minutes very perfect injection of the different tunics may be obtained. The eyes of oxen, from their large size, are most convenient for this purpose, and a pipe may be inserted easily into one of the largest of the small vessels divided in the removal of the eye. I strongly advise the student to inject the eye of the ox, first with the carmine and afterwards with the Prussian blue fluid (p. 105). Beautiful specimens of many of the tissues of the eye may be thus obtained and preserved in glycerine. Small pieces of the prepared eye may be kept in small tubes filled with glycerine, and carefully examined when convenient. In this way I have kept many of the most delicate tissues for twenty years without deterioration. The human eye is not easily injected after its removal from the head in consequence of the small size of the vessels. After an injected eye has been allowed to stand for some time, the different tunics may be examined. In this way beautiful injections of the retina will sometimes be obtained. When eyes are to be preserved for the purpose of subsequent observation, glycerine will be found the most efficient preservative solution, but it should be used diluted with an equal quantity of water in the first instance, otherwise, shrinking to such an extent occurs that the structures are afterwards submitted to examination with great difficulty, whereas if the strength of the glycerine be gradually increased, this inconvenience is avoided.

In order to examine the *choroid*, the sclerotic must be removed, and in doing this it will be found more convenient to cut a narrow strip quite out of the sclerotic in its entire circumference, than to make a simple incision.

410. Of making Sections of the Sclerotic, Cornea, and Retina.—To make sections of the eye exposing the lens and the various tunics *in situ* the eye should be frozen, and a section made with a thin,

sharp, but pretty strong knife. Congelation may be effected by the processes in ordinary use, or by the use of nitrous oxide gas. This is now liquefied or highly condensed in strong iron bottles. As the gas is allowed to issue from a narrow tube inserted into the mouth of the bottle, and fitted with a stop-cock, so much heat is absorbed that not only water, but even mercury, may be very quickly frozen. The degree of congelation can be carefully regulated so that tissues may be only partially frozen if desired. Thin sections of the cornea and retina are made upon the same principle as those of the skin. The sclerotic is first cleaned by cutting away all the muscles adherent to it with sharp scissors, and the eye is then cut into two parts with a sharp knife, without removing the vitreous humour. It may be divided either transversely or in a longitudinal direction. When the cornea is to be examined, the anterior part may be well washed with water, and the ciliary processes, &c., removed; after making little notches around it with scissors, in order that it may dry as flat as possible, it is to be pinned out upon a small piece of board with numerous pins. It is allowed to dry spontaneously, and then thin sections may be made with a sharp scalpel and moistened with water, when they swell out to their former size. The section may be treated with a drop of acetic acid, when the structures of which it is composed will become clearly visible. In order to obtain a section of the retina, the posterior part of the eye with the vitreous humour adhering is carefully notched, and pinned out as in the former case. With care the greater part of the vitreous may be cut away with scissors, but a thin layer should be allowed to dry upon the surface of the retina. Thin sections may be made and treated as in the case of the cornea. Dilute acetic acid or dilute caustic soda may be applied to the section after it has been examined in pure water. The eyes should be perfectly fresh at the time they are pinned out. Specimens prepared in this manner should be compared with others obtained from recent organs. By the methods described very good and instructive specimens may be obtained, but for the study of the distribution of the nerves and of the arrangement of the tissue elements other processes are requisite. The principal of these have been referred to in other parts of this work. The gold process is described on page 70, but for demonstrating the fine branches of the nerves in the cornea, I prefer the cornea of the hyla prepared according to the method described in Chapter VII. In some of my specimens the individual nerve fibres entering the corneal tissue at the margin of the cornea can be followed amongst the bands of fibrous tissue for a long distance. The division and subdivision of the individual fibres can be distinctly seen, and the delicate ramifications can be traced into the ultimate networks of nerve fibres which exist in the very substance of the corneal tissue. In the same specimen the numerous ramifications

of the corneal cavities can be everywhere seen freely anastomosing with the branches extending from neighbouring cavities. In each cavity an oval bioplast exists.

411. Choroid and Ciliary Processes.—Injections of the choroid are made as above described. The greater part of the black pigment may be washed away with a stream of water, or by agitation in water. The ciliary processes, when injected with transparent injection form very beautiful microscopical preparations, and the arrangement of the capillary vessels in the thin layer of the choroid known as the *tapetum lucidum* is most beautiful from its regularity. These specimens may be preserved in glycerine.

412. Examination of the Crystalline Lens.—The crystalline lens may be examined in the recent state by moistening a portion with a drop of water. It may be boiled, and some of the fibres carefully torn off, and afterwards moistened. Or it may be soaked for some time in a solution of chromic acid, and then subjected to examination; or lastly, it may be dried, soaked in oil for a considerable time, and a thick perfectly transparent section made, which may be ground to any required degree of tenuity; the surfaces may be afterwards polished. Sections prepared in this manner may be mounted in Canada balsam.

In examining the character of the fibres of the lens, it is better to boil it previously, and tear off a few fibres with forceps: these may be afterwards carefully separated from each other with needles. The arrangement of the fibres should be examined in different animals, especially in the human subject, the ox, the horse, and in fishes.*

If the disposition of the fibres of the surface is to be shown, the lens requires hardening in a solution of chromic acid. The lens may then be examined in a deep glass cell, in some of the chromic acid solution with a low power.

The development of the fibres of the lens may be studied in the lens of any young animal, which has been properly stained with carmine. The numerous oval bioplasts which take part in the development of the fibres may be very clearly seen.

In examining cataracts we should carefully observe the microscopical characters of the soft external pulpy part, as well as of the hard internal nucleus. In many of these cases numerous oil globules will be observed, which, from my own observations, appear to consist chiefly of cholesterine held in solution in an oily fat; and other larger globules, consisting of some very transparent substance presenting nearly the same refracting powers as this portion of the lens, but evidently of very different composition, as they are not miscible with it, may be observed. Occasionally, also, small plates of cholesterine have been noticed. There is always much granular matter.

* Todd and Bowman's "Physiology," chapter xvii.

Organs of Generation.

The microscopical examination of the generative organs presents no great difficulties, but there are a few points connected with the demonstration of the anatomical characters of some of these which it may be desirable to allude to briefly.

413. Female Organs of Generation.—The *ovary* from its firm, fibrous texture, is cut with some difficulty, and when very thin sections are required, it is better to inject the vessels in the first instance with plain size, or size coloured with transparent injection, in order to give it a consistence more favourable for cutting with a knife. The Graafian vesicles are very readily seen, and their different tissues may be demonstrated by removing small portions with a pair of scissors. The ovary of the bitch or rabbit is more favourable for demonstrating the different anatomical points than the human ovary. In order to examine the distribution of the vessels, it is only necessary to inject the arteries with the carmine or Prussian blue fluid. These organs are highly vascular, and the distribution of the capillaries to the walls of the Graafian follicles is very beautiful.

The *Fallopian* tube may be examined in the same specimen as the ovaries, after the vessels have been injected. The epithelium lining the Fallopian tubes is columnar and ciliated. Its characters are easily made out by examining some of the mucus scraped from the lining membrane of a tube which has been slit up. The arrangement of the vessels of the tube is very beautiful, and thin sections, through the different coats should be obtained, and portions of mucous membrane should be carefully dissected off from the muscular coat, in order that the arrangement of the vessels upon the surface may be examined.

Uterus.—The examination of the uterus should be conducted in the same way. The mucous membrane of an impregnated uterus, which constitutes in fact the *membrana decidua*, is very highly vascular, and the numerous glands which it contains are better displayed in injected than in uninjected preparations. I have made beautiful preparations of these glands without difficulty in the unimpregnated uterus of many of the lower animals as the pig, bitch, cat, and some others. For all these injections I recommend the Prussian blue fluid, and glycerine as the medium for preserving them in.

The mode of demonstrating the character of the muscular fibre cells of the uterus has been already adverted to. The character of these is very different in the unimpregnated organ and at various periods of pregnancy.

414. Male Organs of Generation.—The anatomy of the *testis* is very readily investigated. A portion of one of the seminal tubules is easily drawn out with the aid of forceps and may be examined in fluid.

The pressure of the thin glass must be very carefully avoided. A little solution of caustic soda renders the cells in the interior more distinct. In man, spermatozoa are never found in the seminal tubules, as the development of these bodies is not complete until the cells in which they are formed have arrived at the epididymis. The structure of the testicle is more easily made out in the lower animals. Some specimens in which the vessels have been injected should also be submitted to examination. The testicles of rodent animals (particularly the rat, rabbit, white mouse) afford very demonstrative specimens.

The *Vas deferens* is so hard that thin transverse and longitudinal sections are easily cut with a sharp knife. The fibres of organic muscle in the coats of the tube are demonstrated by the application of a little caustic soda. The characters of spermatozoa are described in the chapter on "Urine."

The arrangement of the vessels of *the penis* should be studied in specimens in which the arteries have been filled with transparent injection and the veins with plain size, or the method of preparation advocated in Chapter VII may be employed. The mode of examining the mucous membrane of the urethra is the same as recommended for the examination of other mucous membranes.

Embryonic Tissues.

415. Investigation of the Structures connected with the Embryo and the Development of Organs.—The highly vascular *placenta* should be studied in organs in which the vessels have been injected first with carmine fluid and then with size and glycerine. The growing bioplasts at the summit of the tufts and the vascular loop growing in their wake as it were, are easily made out in pieces removed from a healthy placenta about the fifth or sixth month and when the bioplasts have been properly stained, the most beautiful and instructive preparations result.

The structure of the walls of the vessels and the intervening tissue of the umbilical cord is well worthy of careful study. The cord should be perfectly fresh and small pieces should be stained in the carmine fluid, hardened in glycerine and chromic acid and preserved in glycerine. Pl. LXXII, fig. 4, p. 464.

The subject of development is a difficult one for the student, and should only be undertaken by those who have much time at their disposal. Advantage will be derived from the use of fluids for hardening tissues previously alluded to, and glycerine will be found to render some structures very distinct, while on the other hand, some tissues are not to be distinguished at all when immersed in this medium. The reader is referred to the remarks on rendering soft tissues hard and transparent, page 45.

I have prepared some beautiful specimens of the developing tissues of the human embryo according to the plan given in Chapter VII. The bioplasts are thoroughly stained and the tissues of an embryo at the very early period of the third week that had been kept in glycerine with a few drops of chromic acid fluid were sufficiently hard to tear into shreds. Some of the developing muscular fibres were very beautiful and could be subjected to examination under the one-fiftieth of an inch object glass without difficulty. Embryos about the eighth week have been well injected with Prussian blue fluid, and the smallest vessels have been satisfactorily distended without rupture and without disturbing the embryo blood-corpuscles that had collected in their interior. The soft pulpy tissues of the embryo improve after they have been preserved for some months in glycerine, and after years have passed an embryo that has been kept in glycerine may be dissected and subjected to minute investigation with great advantage.

The student should obtain embryos varying in age from some of the lower animals, and prepare them according to the plan indicated, and study the development of the several tissues and organs one by one. In this way he will learn many important things concerning development, not to be ascertained in any other way.

In page 48 I have described the method of preparing embryos for studying the development of bones, which was employed by me in 1853. In order to investigate the very early changes in the development of some organs, the heart for instance, it is necessary to harden the developing tissues, which it need scarcely be said must be perfectly fresh, immediately after their removal from the embryo. There are various methods of effecting this object. Among the best is careful immersion in alcohol. I cannot do better than call attention to the directions given by Dr. Tonge in his valuable memoir "On the Development of the Semilunar Valves in the Heart of the Chick," published in the Phil. Trans. for 1869, vol. clix, page 387 :—

"The embryo immediately on its removal from the egg was immersed in strong spirits of wine, the sac of the amnion being first slit open, where this had closed up, in order that the spirit might come freely into contact with every part of the surface of the embryo. As the spirit permeates the tissues of the embryo, which it does rapidly, the capillaries and smaller vessels are soonest contracted by it, and the blood being thus prevented from escaping from the larger arteries before the heart's action is stopped, they become greatly distended. When this distension with blood reaches its maximum in the arteries, the blood accumulates in the cavities of the heart and in the large veins, the impediment caused by the closure of the systemic capillaries and small arteries acting backwards throughout the circulation. By this treatment it was found that the cavities of the heart and the large

vessels nearest to the heart, became fully distended with blood, and becoming coagulated and hardened by the spirit retained the distended condition. The embryos were allowed to remain in the alcohol for a few hours or days, according to their size, in order that they might become completely hardened.

"By careful management of the position of the embryo in the corked glass tube, I was frequently able to obtain clot in the large vessels, owing to the sinking of the corpuscles before the blood had coagulated, or the spirit had penetrated the walls of the artery. The best position for obtaining the result was to keep the head of the embryo downwards, and the body a little inclined towards its dorsal surface. The embryos, after being hardened in spirit, were, of course, perfectly opaque, and could only be examined by reflected light till they had been rendered transparent. This was effected by soaking them in strong Price's glycerine for several days. After this the portions required for examination were removed, and dissected, and examined in glycerine by strong transmitted light under a compound microscope fitted with an erector and a low power (1 inch). The hardness of the tissues permitted sections to be made readily in various places, and their transparency under strong light enabled the course of the vessels distended with coagulated blood to be readily followed. Where it was required to examine the structure of the interior of the cavities of the heart and great vessels, the coagula, if opaque, were first carefully picked out by needles; if a colourless coagulum had been obtained, this troublesome process was often unnecessary. After dissection in strong glycerine the specimens were mounted for drawing and description. They were preserved in glycerine jelly, this medium retaining the specimen in any position in which it was required to be drawn or described, and allowing of its being readily altered to a fresh position when it was necessary that the same object should be seen from different points of view. The observations were made on more than fifty embryos in various stages of development, and the most satisfactory dissections were converted into permanent preparations."

CHAPTER XIX.

The Methods of Examining Morbid Growths and of their General Nature.—*General Characters of Morbid Growths.—Considerations bearing upon the question of the Nature and Origin of Morbid Growths.—Of the Germs of Morbid Growths.—Of extirpating Morbid Growths.—Structure of Morbid Growths.—Fibrous Tumours.—Cartilaginous, Bony, and Myeloid Growths.—Fatty Tumours.—Vascular Tumours.—Adenoid Growths.—Adenoma.—Lymphadenoma.—Myxoma.—Cystic Growths.—Colloid Tumours.—Cholesteatoma.—Recurring Fibroid Tumours.—Epithelial Growths.—Melanoid Tumours.—Fungus Hæmatodes.—Cancers.—Examination of Morbid Growths.*

The elementary structures which may be met with in morbid growths have been already referred to; they are of the same order as those which enter into the formation of normal textures:—granules, globules, cells, fibres, membrane, tubes. Formerly, pathologists considered it of the first importance to give a definite name to every morbid growth, but since the minute anatomy of these structures has been carefully investigated, many of the received names have been shown to be inappropriate. In these days we endeavour to simplify the nomenclature as much as possible, and attach more importance to the accurate description of the structure of a tumour than to the question of determining the name to be given to it. Many of the names handed down to us, and still in use, are objectionable. Such terms as *encephaloid*, *colloid*, *amyloid*, *fibroid* may be applied to a number of structures very different as regards the history of their life and progress, the results to which they lead, as well as their minute structure and composition. There can be no harm in saying a growth has a consistence and colour like *brain* or *gum*, or resembles cells found in the medullary cavity of bones, or that it has a *fibrous appearance*—because we only refer to one of its characters, and there may be many growths agreeing in this, although they differ widely in other essential particulars. But if we call a tumour an *encephaloma*, or a *colloid cancer*, &c., we speak of it as a definite structure which ought to agree in all important characters and vital endowments, with other tumours to which the same name is applied, and with these only. So

far, however, from this being the case, we find tumours may agree in general resemblance, say, to brain matter when examined by the unaided eye, but may differ remarkably from one another as regards all *essential* particulars. It is therefore necessary to study the minute structure of a tumour, and ascertain as far as possible, its history, instead of merely attempting to assign to it a name, such as, *scirrhus*, *fibrous sarcoma*, &c. There are many tumours which have been called *scirrhus* that were merely *fibrous*, and *vice versâ*. The slovenly methods of observation which have been adopted even by some who have been *most* rigid in classifying and naming tumours, have led to very erroneous conclusions.

As we learn more concerning the anatomy, mode of development, and history of morbid growths, we become less disposed to attempt anything like a systematic classification, and it is much more desirable to give good drawings of the structure of the growths and a short description of their most important characters, than to give them names, still less to hide our ignorance of their real nature by the use of such imposing terms as "*Fibrocystic sarcoma*," "*Cylindroma*," "*Cholesteatoma*," and many others, which merely connote one or two characters, and may include a number of structures which are essentially distinct from one another under the same name. Rather let it be said that a tumour is like brain, or marrow, or suet, or that it has a fibrous, cartilaginous, vascular, glandular, or osseous appearance; or that it contains plates of cholesterine, or is made up of cysts, &c.; or that it is composed of fibrous tissue, epithelium, cancer cells, mucous tissue, a gum-like material, &c. If we do this, any one who examines our work afterwards can form an idea of what we saw, while by merely attaching a name to the structure we simply add to the doubt and confusion which already exist, especially as the meaning of the term we use will, in all probability, be much altered in the course of a few years.

It is not easy to point to special characters by which many of these growths may be grouped together in certain broadly characterised classes. Not only is there a difficulty in defining the different tumours by their microscopical characters, but the so called benign tumours pass by almost imperceptible shades into those of a malignant and dangerous nature. Upon the whole, it will probably be agreed that the safest basis for arrangement is that which is founded upon the alliance between the morbid growth and normal tissue from which it sprang.

Many of the morbid structures referred to in this chapter used to be regarded as new formations. But now we know that every one of these morbid growths begins its life as a particle of bioplasm or as a collection of bioplasts, and we trace this living matter back to its origin in pre-existing living matter. There is, therefore, no such thing as a *new formation* in the strict sense of the term. The development of a

tumour from indifferent or non-living matter is as impossible as is the development of one of the normal textures from lifeless substance. Every morbid growth is connected by direct descent with bioplasm which existed before it.

416. General Characters of Morbid Growths.—Morbid growths and tumours are met with in various parts of the body, and sometimes appear quite superficially ; sometimes they are deeply embedded in the substance of solid organs, such as the liver or brain, and derive their nutriment from every point of the surrounding texture, or are united to the adjacent tissue by a long narrow pedicle containing the necessary vessels and nerves for the supply of the tumour.

A tumour may be produced by the irregular growth of a tissue at a particular point, in which case it consists simply of the elements of this tissue. Fatty tumours, certain tumours of a fibrous structure, exostoses from bones, and many others, are produced in this way, and, as might be expected, but little difference can be made out between their minute structure and that of the tissue of which they are, as it were, the off-growth. In other instances, however, and these are extremely numerous, the morbid growth is found to possess a structure of a different character ; and although in it elements of one or more of the tissues in a healthy state may be discerned, it may differ in important characters and in its rate of growth from every normal texture of the body.

In taking a general survey of the more common morbid growths which are brought under our notice, and examining carefully into the tissues involved, we cannot fail to remark the peculiarly localized condition and limited origin of many of them. Often an enormous mass appears to have been formed by the rapid and circumscribed growth of one or more elements in one particular spot in a tissue. By a redundant growth of epithelium on some part of the cutaneous surface, a corn may be produced ;—by simple hypertrophy of the subcutaneous areolar tissue of a part of the leg or foot, or of that of the scrotum, most formidable diseases are caused ; subcutaneous fibrous tumours depend upon a morbid development of the same tissue, only it is still more circumscribed. The general appearance of hypertrophied areolar tissue is represented in pl. LIX. The specimen from which the drawing was made was taken from the scrotum of a patient operated upon by Sir W. Fergusson. Upon the addition of acetic acid to the preparation, the fibres of the yellow element, fig. 2, pl. LIX, page 380, became very distinct. Such tumours as the above are detrimental principally by their bulk and used to be spoken of as "benign."

The so-called *cancerous* growths which not only grow quickly to a vast size, but may spread to distant parts of the body and may recur if extirpated, frequently commence deep in the substance of a tissue, and

gradually make their way towards the surface. In many instances the tendency to the development of cancerous growths is hereditary. As Mr. Moore has remarked, they often occur in persons who have exhibited remarkable health and vigour of constitution. Sometimes they appear to spring up in different and very distant parts of the body at the same time. The cells, of which these tumours are in great part composed, possess a very remarkable power of multiplication, and it has been truly said that if even a little of the '*fluid*' they contain be carried to distant parts of the body, it may give rise to the development of germs which will become tumours, and encroach upon the structure in which they may have taken root. Schroeder van der Kolk held that from the fluid of a morbid growth cells might be developed, which might increase until a tumour like the original one was formed. Dr. Bennett entertained a similar opinion. But we now know that the '*fluid*' contains millions of germs or minute living bioplasts, every one of which might grow under favourable conditions.

The cancerous tumours thus originating usually resemble the first one in their essential points of structure, but differ from it according to the nature of the tissue which has been invaded. If, for instance, the growth takes place in a part where areolar tissue is abundant, and where there is considerable resistance to its increase, we may expect to find a hard, condensed, and fibrous tumour; but if the growth commences immediately beneath the surface of the peritoneum, or in a like situation, where it will encounter little resistance, a soft, spongy structure will probably be formed, in which the cells preponderate over the fibrous element.

Considerations bearing upon the question of the real Nature and Origin of Morbid Growths.

It may be considered as a fact beyond dispute that every healthy and morbid tissue formed under any circumstances in living beings results from changes taking place in bioplasm; and that this bioplasm whatever its properties or powers may be, came from bioplasm which existed before it, possessing in some cases similar, in others very different, properties or powers. The bioplasm of every texture in the body is derived by continuous descent from the original embryonic germinal mass and every particle of morbid bioplasm must also be regarded as a direct descendant of this.

It is interesting to contemplate the extremely complex but orderly phenomena which result in the formation of a normal tissue. The multiplication of the masses of bioplasm, the production of formed material, the supply of pabulum, the removal of the substances resulting from chemical changes which take place, must all proceed with regularity, and in perfect order, for otherwise the resulting tissue

will not be normal. Even if the supply of nutriment be modified in quantity or quality, a difference in the character of the tissue or organ produced will be manifested.

It has been truly remarked that many morbid growths could not easily be distinguished in their ultimate structure from the healthy tissue. In fact the only difference in some instances seems to be that the growth of the latter is regular, even, and restricted; while the former grows irregularly as regards the rate of increase, unevenly as regards its form, and there seems to be no limit to the size it may attain, if it be freely supplied with nutrient material. Whatever may be the nature of those complex conditions to which the symmetry of the body is due and which necessitate or enforce a symmetrical arrangement and definiteness of form of the various parts of which the organism is composed, they must be absent in the case of some of the simplest morbid growths. Many of these are supplied with vessels which could not be distinguished from those of healthy tissue. The arteries, capillaries, and veins exhibit precisely the same structure; the arrangement of the muscular fibres of the walls of the first and last channels is precisely the same as that met with in healthy vessels, and there can be little doubt that they are supplied with nerve fibres upon the same plan. But it is not impossible, indeed it seems very probable, that the regular growth and destruction which are so remarkable in every part of the nervous system in the normal condition are departed from in the case of the nerve centres and of the nerve fibres which are distributed to morbid growths.

Now there is one system of vessels intimately connected with the blood vessels, the importance of which in vertebrate animals is far greater than has yet been supposed, of the arrangement of which in morbid growths nothing is yet known; and its existence in many, at least in its normal characters, is extremely doubtful. We know that in health *lymphatics* are freely distributed upon the surface of those elementary organs of which the lungs, the liver, muscles, &c., may be said to be made up. And we know that these lymphatics contain a fluid in which particles of living bioplasm, capable of taking up various materials resulting from the decay of tissues, are suspended. It seems at least probable that the lymphatics may have much to do with maintaining regularity of growth, and of preventing a redundant production of tissue. And it would therefore be extremely interesting if we could obtain an accurate knowledge of the distribution of these vessels in morbid growths.

Whatever may be the circumstances which lead to the production of a morbid growth there can be little doubt concerning the general nature of the texture of which it consists. Nor can there be a question that the very bioplasm from which this so-called adventitious growth

has proceeded, originally sprang from the same living matter which gave origin to that which took part in the formation of the normal tissues of the body. In some cases in which the abnormal is directly continuous with the normal structure, it is impossible to define the limits of either, and to state exactly where the morbid tissue commenced or the normal one ceased, so gradual is the passage from the normal to the abnormal.

The *fully-formed* anatomical elements of a normal tissue could not give origin to a morbid growth. We know, however, that in all normal textures are collections of small masses of embryonic bioplasm from which new texture is from time to time formed, to take the place of that which is gradually destroyed in the performance of its function, and by which, in some cases, portions of new tissue may be developed, if that existing is called upon to do an increased amount of work, or where a portion is destroyed by disease. Such embryonic masses are to be demonstrated in all growing tissues at every period of life, and are most abundant in those textures which grow continuously. The rate of the growth and development of these bioplasts varies according to the rate of destruction and removal of the textures which are to be replaced. And this must vary extremely at different periods of life and under different circumstances; thus almost any tissue or organ in a man's body may at one time of life be considerably reduced in volume, and the amount of work performed by it greatly diminished, while at another it may be greatly increased in size, and the quantity of work performed by it may be doubled or trebled. And there is reason to think that the process may be repeated more than once at different periods of life if the organism is a healthy one. Now, suppose that one such embryonic mass should be irregularly and abundantly supplied with nutrient material, and sprout, as it were, into active growth when it was not required,—a shapeless lump of tissue would result, partaking of the characters of the normal tissue, and apparently continuous with it,—in fact, a small tumour which would continue to grow by the formation of new bioplasm in precisely the same manner as the normal texture is developed, except that the conditions which regulate and limit growth, and preside over the symmetry of the texture formed, are absent, or their influence counteracted.

But of these masses of embryonic bioplasm there are two kinds:—

1. Those which are concerned in the development of the individual tissues.

2. Those from which complex tissues or elementary organs, including vessels and nerve fibres, may be developed.

The first are found in close relation with the respective tissues they represent and cannot be missed by the observer who studies properly prepared specimens, but the last are met with in the substance of

complex tissues and organs, and may be passed over unless the greatest care be taken, and, indeed, they are very difficult to demonstrate. They exist, however, in connection with all complex organs of the body, and are to be found in the muscles and nerves, in the kidney, liver, and other glands, not only in the child but in adult life, and even in old age.

If a part of an organ be destroyed by injury or disease, new structure may be developed from these masses of embryonic bioplasm. Even in man there is evidence of this extension of embryonic developmental power far into life, but in many of the lower animals, as is well known, it exists to such a degree that complete and perfect limbs or organs may be developed anew in the fully formed animal after the removal of those produced in the course of development and growth.

One or other form of these embryonic masses of bioplasm connected with the normal tissues, invariably, I believe, constitutes the source or origin, or is the starting point of a morbid growth. And it seems not unreasonable to suppose that by a mechanical injury, or in consequence of changes proceeding in tissues in the immediate neighbourhood, the position of one or more of the masses of embryonic bioplasm may be somewhat altered in its position and relations. In consequence of being more abundantly supplied with nutrient material in this new situation, they would grow out of their proper order and perhaps very quickly. Such irregular and increased growth might commence at almost any period of life, and might be determined by change in the distribution of nutrient matter, by irregularity of development consequent upon changes commencing perhaps at an early period of intra-uterine life, or by important changes having taken place in the arrangements connected with the processes of destruction and removal of tissue or in the mechanism which regulates and governs these phenomena.

As we find that the masses of bioplasm resulting from the original embryonic mass, and continuously descended from it, exhibit very different properties and powers from those of the original mass itself, it is not surprising that masses of living matter resulting under the influence of unusual conditions and altered pabulum, should manifest powers which the original mass from which they proceeded did not possess. And as regards the degree of departure from the normal type, I would remark that this would be far more marked in the case of the tumour developed from the second form of embryonic bioplasm than in that from the first, by reason of the far greater developmental capacity possessed by the bioplasm which most closely accords with that which exists in an early period of embryonic life.

For the acquirement of the utmost symmetry and perfection which it is possible for the normal organism to attain, the multiplication and development of the several masses of bioplasm must proceed with

regularity. No wonder then that the irregular nutrition and too rapid multiplication of such masses result in the production of a chaotic texture which serves no useful purpose in itself, and may ultimately lead to the destruction of the organism in connection with which it is developed. With regard to the precise cause of the regularity occurring in normal growth and development, our knowledge is still imperfect. It was shown on page 248, that under altered conditions the bioplasm of an elementary part, or cell of epithelium would give origin to masses differing extremely in property from the original mass. Its descendants could never again form epithelium, or probably perform any office advantageous to the individual, though in comparison they would grow and multiply much faster than the original mass, and would retain their vitality under circumstances which would have led to the destruction of the first. It is therefore only in accordance with facts which I think may be relied upon, to assume that where the altered conditions affect not only the elementary parts already formed, although young, but those from which vessels and those from which tissues are to be developed, these latter should possess structural characters and properties, which are wanting in the normal tissue, or at any rate while it remains under the ordinary conditions.

If the embryonic masses of certain normal tissues be transplanted from one part of the body to another, or even be removed to a different organism, they will grow, and a new texture precisely corresponding to that from which the germs were taken will be produced in the new situation. When the multiplication of bioplasm has gone on with undue rapidity, the resulting masses acquire increased powers of living under varying conditions. Under certain altered conditions masses of bioplasm resulting from these embryonic masses may acquire such exaggerated powers of growth as to move about freely, absorb nutriment faster than usual, and multiply with great rapidity. Insinuating themselves between the elements of the normal fully-formed texture, they may grow and multiply at its expense, and, of course, may lead to its destruction. The faster they multiply the more independent they become. They may pass into lymphatic vessels or into the blood, or they may travel long distances, like the germs of entozoa, until having arrived in a locality where they are abundantly supplied with nutrient material, they may grow and establish growths in distant parts, resembling at least in important particulars—that which was first produced. Such masses of bioplasm also live and grow and multiply at the expense of the very formed material they have produced.

That tumours should differ materially in the rate at which they grow is exactly what we should expect when we consider how the normal tissues of the body differ from one another in this particular. A growth allied in its character to fibrous tissue, will increase much more slowly

than one which partakes of the characters of epithelium, or some other more rapidly-growing tissue. But the morbid growths which correspond to the tissues which grow most slowly, increase very rapidly if the rate is compared with that of the former; and it is commonly observed that the power of rapid growth increases as the multiplication of the elementary part proceeds. Thus the elements of a bony tumour which grows very slowly at first may multiply with wonderful rapidity after the growth has attained a certain size. As the rate of growth increases the characters of the elementary parts become modified, and the proportion of the bioplasm to the formed material is considerably greater in a quickly than in a slowly growing cell.

Anything interfering with the regular growth of a continuously growing structure in the adult, may ultimately lead to the formation of a tumour more or less closely allied to cancer; thus, suppose there be any impediment to the escape of the growing hair from the surface of the skin, growth would continue from the bulb, and the modified hair produced might be caused to assume the form of a compressed spiral, and this, with the changes necessarily resulting in adjacent textures would soon give rise to the formation of a considerable tumour, exhibiting a very complex structure. If the regular growth of cuticular epithelium be interfered with in a certain way, a morbid growth which may assume the characters of what is known as epithelial cancer may result. The state of things which brings about this change usually soon disturbs the relative position of the papillæ, upon the surface of which the cuticular cells are formed. Many of these may be made to grow towards one another, so that there is no escape for the modified epithelium which is produced. It would seem that as this irregular growth of epithelium and of papillæ with the modified connective tissue, vessels and nerves of which these are composed, proceeds, the new properties of the elementary parts are acquired, and a tumour results, the structure of which differs much from that of skin, but in which, nevertheless, bodies representing all the normal anatomical elements of the cutis and cuticle can be detected without difficulty. That some growths (cancerous) should manifest extraordinary powers of multiplication and distinctly specific characters which they retain if transplanted, is very remarkable—but perhaps not more so than the fact that pus developed on the surface of the peritoneum acquires peculiar specific poisonous properties not belonging to other forms of pus.

While I must admit my inability to draw sharp lines of demarcation between different kinds of morbid growths, and that the same growth may exhibit not only very different anatomical characters, but manifest diverse properties or powers at different periods of its existence, and am unable to suggest any satisfactory classification of these structures,

I venture to think they may be conveniently arranged in some such groups as suggested in the following divisions :—

1. Morbid growths which are due to an irregular growth of a simple tissue without any or with very slight alteration in structure; such are corns, certain fibrous and cartilaginous and bony growths.

2. Those which have originated in normal structures or organs of the healthy body, but the anatomical elements of which are much altered.

3. Those which consist of the several anatomical elements of a complex tissue—more or less modified in structure. Among these would be placed fatty tumours, some glandular or adenoid growths, and some others.

4. Morbid growths (malignant, cancerous) originating in the embryonic bioplasm found in connection with complex structures, and exhibiting in their fully developed state a very wide departure from any healthy tissue—not only in structural characters but in vital characteristics. Such growths exhibit the widest departure from any structures of a normal type.

Of the Germs of Morbid Growths which may be transmitted in the Blood or Lymph to different parts of the Body.

The particles which are capable of giving rise to a morbid growth may be extremely minute—so small that they could readily pass through the walls of a capillary vessel. Many observations render it certain that insoluble particles possessing marvellous powers of development are far more minute than used to be supposed. And there is no doubt that powers and properties which were formerly attributed to fluids, are really due to the insoluble particles of living matter suspended in the fluid. Just as a minute particle of a living pus corpuscle, pages 125, 305, if transplanted to a new soil suitable to it, may grow and give rise to new pus corpuscles, having precisely the same properties; so, the most minute germ of a cancer cell may, under favourable circumstances, give rise to the production of cell forms exactly resembling those in the original tumour. [1865.]

Of Extirpating Morbid Growths.

The germs of the most fatal varieties of cancer are remarkable for their rapid multiplication and for the power they exhibit of resisting the influence of adverse external conditions. Not only do they invade, destroy, and live at the expense of normal textures, but they travel for such considerable distances and spread so far from the focus of their original development that after a tumour has been growing for some time it is almost impossible to extirpate it. And although a considerable portion of surrounding healthy textures may be removed with it,

it but too often happens that particles of the cancerous bioplasm far too minute to be detected by ordinary examination remain behind, and soon grow and multiply, leading to the formation of new masses like the parent growth. Various remedies which have the property of destroying living tissues have been employed for the purpose of extirpating these growths more effectually than was possible by the knife. Of these substances probably a solution of chloride of zinc (about thirty grains to the ounce) is by far the most effectual. The very valuable observations of the late Mr. Campbell de Morgan, at the Middlesex Hospital, conclusively proved the advantages of this plan of treatment in many cases. Moreover, what we know of the nature of abnormal, rapidly-growing bioplasm, really justifies the hope that ere long remedies may be discovered which will destroy it without acting deleteriously upon the adjacent normal textures. The highly interesting observations of Mr. Crookes proved conclusively that carbolic acid would destroy the life of certain living particles while others resisted its influence. And the evidence adduced by me from a totally different method of enquiry went far to prove that the bioplasm of a contagious disease differed remarkably in its vital properties and powers from that of a healthy tissue. Now, since there are certain vapours and solutions which will destroy the life of rapidly growing particles such as pus and those which as I believe constitute the virus of contagious diseases while they do not affect the normal bioplasm of the organisms of man and the higher animals, protected as this is for the most part by its formed material,—it seems not unreasonable to anticipate that a destructive material may some day be discovered and employed of such strength as to destroy cancer cells and leave the normal tissue and its bioplasm intact. It seems even possible that something of this kind might be introduced into the blood, and while circulating through the system, extirpate the germs of rapidly growing morbid structures. At any rate the considerations above very imperfectly sketched clearly indicate the importance of the more minute investigation of the structure of morbid growths, and the more thorough and careful study of the conditions under which they originate and grow. So far from such minute enquiry being really opposed to common sense, it is likely to lead to practical results, and may enable us to answer questions that would suggest themselves to any sensible person with reference to the origin, nature, prevention, or cure of these diseases.

General Characters of some of the most important Morbid Growths.

In examining morbid growths the student should take special note of *a*, the arrangement of the fibres or the fibrous structure; *b*, the number and mode of distribution of the vessels; *c*, the form, number,

and arrangement of any bioplasts or cell-like bodies,—particularly the manner in which they grow, and the situation of the youngest and oldest bioplasts, and the thickness, hardness, transparency, or otherwise of any formed material that may exist around the bioplasts or in which they may be embedded ; and lastly, *d*, the characters and proportion of the fluid or viscid matter which occupies the meshes of the growth—or which intervenes between the cells or bioplasts. The presence of other anatomical elements as *bone*, *adipose tissue*, *&c.*, should be noted. The use of such terms as *fibro-sarcomatous*, *fibro-cystic sarcoma* should be avoided, but an accurate description of what has been seen should be given, and careful drawings should be appended.

417. Fibrous Tumours.—A vast number of morbid structures may be described as *fibrous*, but many of these differ very much from one another in important characters, as for instance in mode of origin, rapidity of growth, and minute structure. Some are composed of exceedingly delicate fibres, others of wide fibrous bands having distinct bioplasts scattered in them. In some a number of minute elongated cells may be detected, while others seem to be composed of fibres with oval masses of bioplasm situated at tolerably regular intervals. Much difference in structure is often observed in different parts of the same tumour. In many cases this is to be ascribed to difference in age.

Fibrous tumours may be connected with the skin, mucous membrane, glands, muscle, nerve, bone, cartilage, and other textures. Some are exceedingly soft, and consist of a delicate network of fibrous tissue containing a soft albuminous material in its meshes. (*Myxomatous growths*, p. 461.) Others are almost of a cartilaginous consistence, and not a few contain bone. Fig. 1, pl. LXXI, is an example of a rare form of fibrous tumour, in which the bioplasm is very abundant. It was removed from the tongue of a patient, and had been growing about two years. It was painless, and very slowly increased to the size of a pea. This specimen was sent to me by my friend Dr. Eade, of Norwich, fig. 1, pl. LXXI.

Involuntary muscle often undergoes change, forming a firm, hard, fibrous-like tumour. A section of such a mass is represented in fig. 2, pl. LXXI.

Cartilaginous, *bony*, *and myeloid growths* have been already referred to in pp. 376, 377. Drawings of the so-called myeloid cells from healthy bone and from a tumour are figured in pl. LXXI, figs. 3 and 4. An interesting specimen of altered osseous growth is represented in fig. 5, for which I am indebted to my friend Prof. Brown.

418. Recurring Fibroid Tumours.—This term has been applied by Sir James Paget to a form of fibrous tumour which returns after extirpation. These tumours are hard and firm, and consist of elongated cells and long fibres prolonged from small cells arranged in an arched

manner. Sir James Paget has remarked that when *new* fibrous growths are formed after removal, they exhibit a greater resemblance to truly cancerous tumours than the original growth.

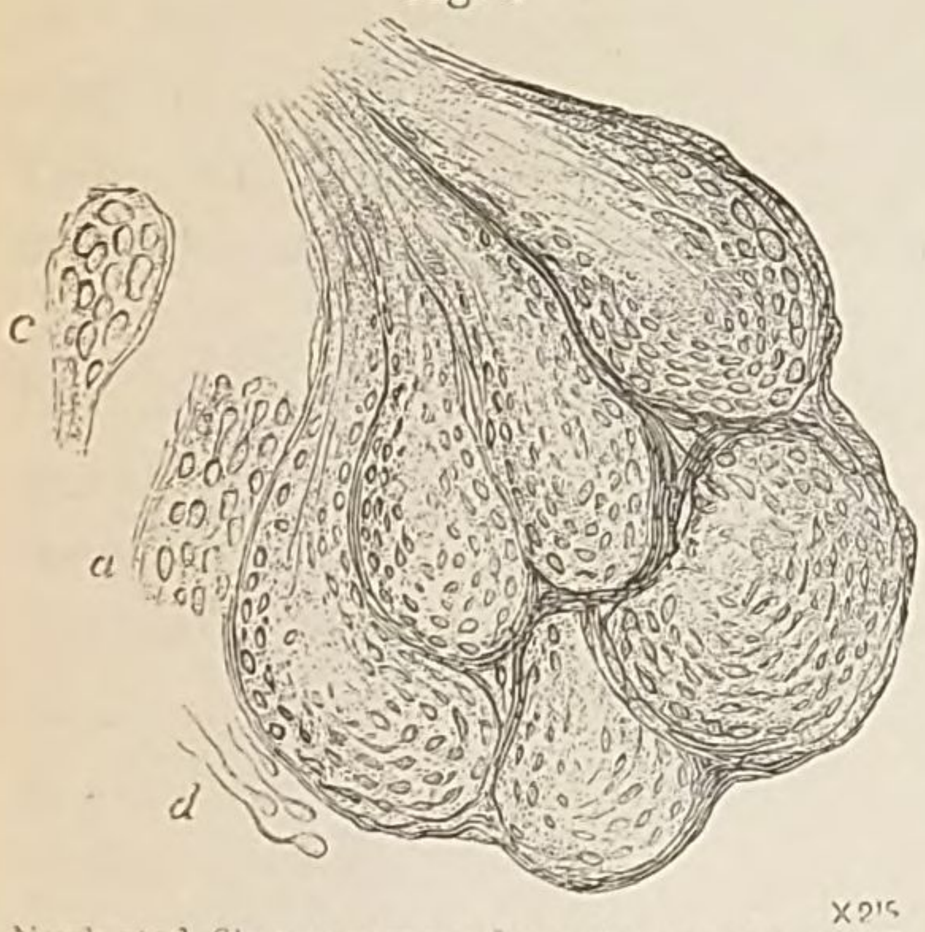
Fig. 1, pl. LXXII, is an example of a tumour probably of this nature. It was removed from the testicle of a man aged sixty. It was as large as the fist, and the testicle was adherent to its lower and outer part, but was not contained in it. For the specimen I am indebted to my friend Dr. Eade, of Norwich.

419. Phlebolithes and Brain-Sand.—The first are hard rounded bodies which are not uncommonly found in veins. They are more common in the veins of the pelvis than in those of other parts. Sometimes the vein is obliterated and the concretion appears to be connected to adjacent parts merely by a pedicle. They consist of phosphate and carbonate of lime with animal matter. The materials are deposited in successive layers upon the lining membrane of the vein, with organic matter; the most internal layers being the oldest, contain the largest quantity of inorganic material. Calcareous matter is sometimes deposited in the smaller bronchial tubes and in cavities of the lung in the course of phthisis. See p. 279, pl. XXXIX, fig. 3, p. 276.

The so-called *brain-sand* consists of rounded masses of carbonate and phosphate of lime. See pl. XXVI, fig. 2, p. 230. The earthy salt is deposited in an organic matrix and previous to its deposition the transparent spherical mass very closely resembles a starch globule, and for this reason has been termed an *amyloid body or corpuscle.* It is composed of albuminous matter, not starch, and though iodine produces a slight reaction the change cannot be compared with that which is well known to indicate the presence of actual starchy matter.

420. Fatty Tumours have a structure resembling that of ordinary adipose tissue. They are often found in connection with the normal fatty tissue of the body. Some of them contain a considerable quantity of fibrous tissue. The subcutaneous adipose tissue, especially of the nose, is liable to increase considerably in quantity, producing horrible deformity. This condition is termed lipoma. Fig. 3, pl. LIX, page 380, shows the structure of a large fatty tumour connected with the testicle, which was removed by Sir W. Fergusson. This tumour was as large as the head, and was in part fibrous and partly fatty. In fig. 6, ordinary adipose tissue with its capillaries is shown. After fatty tumours have been preserved for some time, crystals of *margarine* form upon the surface of the oily fat, as represented in the drawing, fig. 3. Some fatty tumours which contain a quantity of fibrous tissue as well as adipose vesicles, are termed *steatomatous.* Steatoma is also applied to encysted tumours originating in the sebaceous follicles, and containing a soft, pulpy material, rich in fatty matter, but not containing fat vesicles. The fat is partly in the form of small globules,

Fig. 1



×215

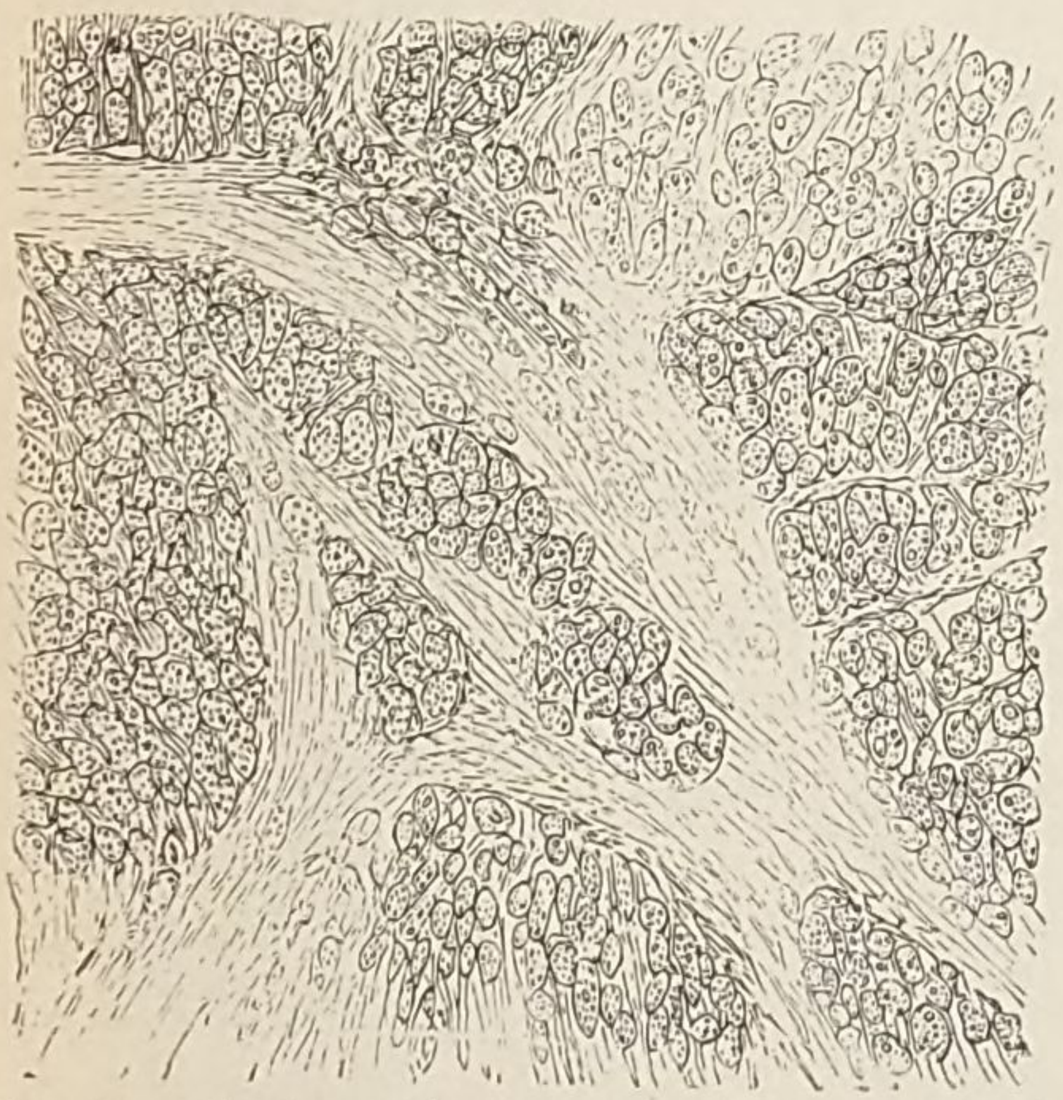
Nucleated fibrous tumour from the tongue. × 215. *a*, part where the cells are very distinct. *d*, fibres seen to be continuous with the cells *c*, small lobule only containing a few cells, and showing the manner in which the tumour grows. M.B., 643 p. 459.

Fig. 3.

×300

Cancellated structure of bone and myeloid cells. Human. *a*, fully formed bone. *b*, capillary vessel. *c*, fibrous or connective tissue. *d*, one of the so-called myeloid cells. *e*, another myeloid cell, forming a curve round the capillary. p. 377.

Fig. 2.



Section of the thickened pylorus, showing bands of pale muscular fibres cut across. × 215. p. 463.

Fig. 4.

Myeloid cells from a soft tumour at the lower end of the shaft of the femur. × 220. From a drawing by Mr. HULKE. p. 459.

Fig. 5.

Transverse section of the meta-tarsal bone of a goat, affected with mollities ossium, *a*, cancellated structure. *b*, enlarged Havessian canal. *c* normal Havessian canal. × 25 Prof. BROWN, "Veterinarian," April, 1869. p. 256.

1000th of an inch ——— × 130.

" " ——— × 215.

To face page 46

partly granular. Sometimes the globules have run together forming large masses of oil.

421. Myxomatous Growths.—Certain soft, mucus-like growths have received this name. They are found to consist principally of stellate or branching cells, which form a soft, delicate, moist, fibrous texture, closely resembling the tissue found between the vessels of the umbilical cord and known as the "mucous tissue" of that structure, fig. 4, pl. LXXII. Many of these morbid growths originate in some form of connective tissue; indeed they may be said to consist of a delicate form of connective tissue in which the yellow element is present in very small quantity or is entirely absent. The rate of growth of some of these tumours is rapid, but they are usually circumscribed and if completely removed do not return. They do not give rise to germs which may pass into the lymphatic or blood vessels and thus be carried to distant parts and develop growths of the same character.

422. Colloid Tumours are soft and jelly-like. They are composed of a viscid albuminous material, held in spaces having fibrous walls, or in the meshes of an exceedingly delicate network of fibrous tissue in which the vessels ramify. On the walls of the spaces are often found a number of roundish cells, some of which are also suspended in the transparent jelly-like contents. In some cases these round or oval cells contain oil globules which are also found free in the fluid.

Colloid Cancer has been applied to tumours of this description which are more rich in cellular elements, and prone to appear in different parts of the body. By some the *ovarian tumour* is considered as a form of *colloid*, but its history and mode of development differ from the gum-like growths met with in other localities. Drawings of good examples of colloid tumours will be found in vol. v. of the "Transactions of the Pathological Society," page 320. The peculiar appearance of these growths is due to the large amount of albumen they contain. I found the composition of one weighing three pounds, removed from the calf of the leg, by Sir W. Fergusson, to be as follows:—

Water	904·60
Solid matter	95·40
Extractive soluble in water	15·20
Albumen	64·20
Fatty matter	4·53
Alkaline salts...	7·60
Earthy salts	2·85
Sulphuric acid	1·05
Phosphoric acid	2·912

The so-called *colloid corpuscles*, are small, round, or oval bodies com-

posed of several layers of a clear transparent substance. They have been described by Hassall, Virchow, Kölliker, and others, and have been termed *corpora amylacea* by some observers. They have no connection with the colloid growths, and are only alluded to here in consequence of the term *colloid* having been applied to them. See p. 460.

423. Vascular Tumours are those which consist principally of small vessels and have been included under the head of *Angioma*. Aneurisms by anastomosis, and certain forms of nævi are of this character. The tumour contains besides vessels a certain quantity of fibrous tissue. Many of these tumours, however, consist principally of veins of considerable size, while some are formed originally of capillaries which undergo considerable dilatation. Vessels in various organs are liable to become varicose, and sometimes irregular dilatations are met with in the brain, retina, and in glandular organs. In such instances, the most important structural elements of the tissue may be obliterated and the growth may partake of the characters of Angioma. Thus cancerous tumours often become highly vascular. The so-called *Fungus hæmatodes* is a malignant tumour infiltrated with blood and containing a number of gorged vessels. The presence of the cancer cells, however, if these be well marked, at once determines the nature of the tumour. In some of these vascular growths there can be little doubt that the vessels are developed in the structure itself, from cells, as in embryonic tissues. When an opportunity offers of investigating the structure of these growths, the tumour should be injected with size and glycerine slightly tinted with Prussian blue injection.

424. Glandular Tumours. Adenoma. Adenoid Tissue. Any glandular structure may exhibit irregular growth until a tumour of considerable size may result, the structural characters of which will vary according to the particular glandular structure involved. The tubes of a tubular gland, the kidney, for example, may give off diverticula here and there, and these may continue to grow and form irregular tubes or sacs containing modified epithelium. Sometimes closed cavities result and the formation of these and portions of tubes, with of course vessels, some nerve fibres, and a certain amount of connective tissue may continue until a quantity of morbid structure, incapable of functional action and quite useless to the organism, results.

Polypi of mucous membrane of nose and uterus are usually regarded as adenoid growths, but although some of these remarkable morbid growths do exhibit glandular elements and contain cysts formed by the obstruction and dilatation of some of the secreting tubes, others contain no vestige of glandular elements, and it is doubtful if they originate in, or represent any epithelial or secreting glandular structure.

425. Nerve Tumours.—Of nerve tumours there are many kinds which differ in structure according to the particular elements of the

nerve tissue principally involved. Some originate in the bioplasm of the nerve fibres, others in that of the nerve cells and some begin in the bioplasm of the connective tissue of nerve structures. Around such tumours, the nerve tissue is often found altered and disintegrated from stretching or pressure, as it is sometimes in the neighbourhood of a clot, fig. 2, pl. LXXII.

A tumour which consists of small bioplasts with a little very fine connective tissue amongst them has been called glioma. It is supposed to originate in the connective tissue, but more probably it is developed from bioplasts, which, at a later period of life, might have become nerve cells. It is in the large central nerve organs that such tumours are not uncommonly found. They do not infect the system and appear in other parts.

Very interesting are the tumours which result from a developmental modification of the so-called caudate cells of the gray matter of the brain spinal cord, retina, and other parts, for they retain the peculiar anatomical characters of the cells, though they have lost all power of discharging functions, and constitute a mass detrimental to the organism, and which may seriously impede the action of the cells and tissues adjacent to them, besides perhaps exciting distal irritation. A tumour of the kind referred to has been described and figured in pl. IX, fig. 4, Vol. I, of the Archives of Medicine, p. 52.

426. Muscle Tumours or Myomata originate from the bioplasm of muscle, but the tissue formed is quite unlike that of voluntary muscular fibre and devoid of contractility. The commonest forms of muscle morbid growths are found in connection with the muscular coats of the stomach, and intestines, and the uterus. These all exhibit resemblances to the normal, unstriped muscular fibre, but it need scarcely be said the tissue does not manifest the properties of muscle. It is devoid of contractility, and the growth of the fibres is irregular, and they cross one another at various angles. In many instances the amount of this abnormal, hard, rigid, gristle-like tissue is very great. See page 397. Fig. 2, pl. LXXI, p. 448, represents a section of a good example of a tumour of this description, which was sent to me by Dr. Hall, of Brighton. It was taken from the body of a man who had vomited sarcinæ for a considerable period of time.

427. Cystic Growths are met with in almost all parts of the body. They are produced in many different ways and their contents are various. Some are filled with a perfectly transparent fluid as limpid as water; others with a thick pasty material; and some contain perfectly hard calcareous matter. Cysts may be formed as follows:—

1. *If the duct of a gland be obstructed, and the secretion accumulates behind the occluded point, the tube becomes dilated, and a cyst is at length produced, with probably ultimate destruction of the gland structure.* The

contents of the cyst undergo gradual alteration, and often when examined are found not to resemble in any way the secretion of the gland. In this way cystic tumours connected with the ducts or secreting tubes of the liver, kidney, sebaceous, mammary, salivary, and other glands are formed. The whole kidney has been converted into one large cyst from obstruction of the ureter. Cysts may be developed in the uriniferous tubes in the cortical and medullary portion of the kidney.

2. *By the increase in size of the areolæ or spaces between the structures entering into the formation of different glandular organs.* The small serous cysts in connection with the villi of the placenta, and choroid plexuses of the brain, and certain cysts met with in the liver, kidney, and other glandular organs, are probably formed in this manner.

3. *By the gradual formation of cavities by the degeneration and absorption of portions of the normal structure.* The spaces become occupied with fluid and a smooth wall is gradually formed upon the interior of the cavity. Some cysts which are met with in the brain, liver, and other solid organs are probably formed in this manner.*

4. *By the increase in size of a single cell, the walls of which become thickened by the deposition of new material.* The cavity of the cyst is supposed to correspond to the cavity of the original cell. This view is still maintained by some, but I doubt whether it is correct.

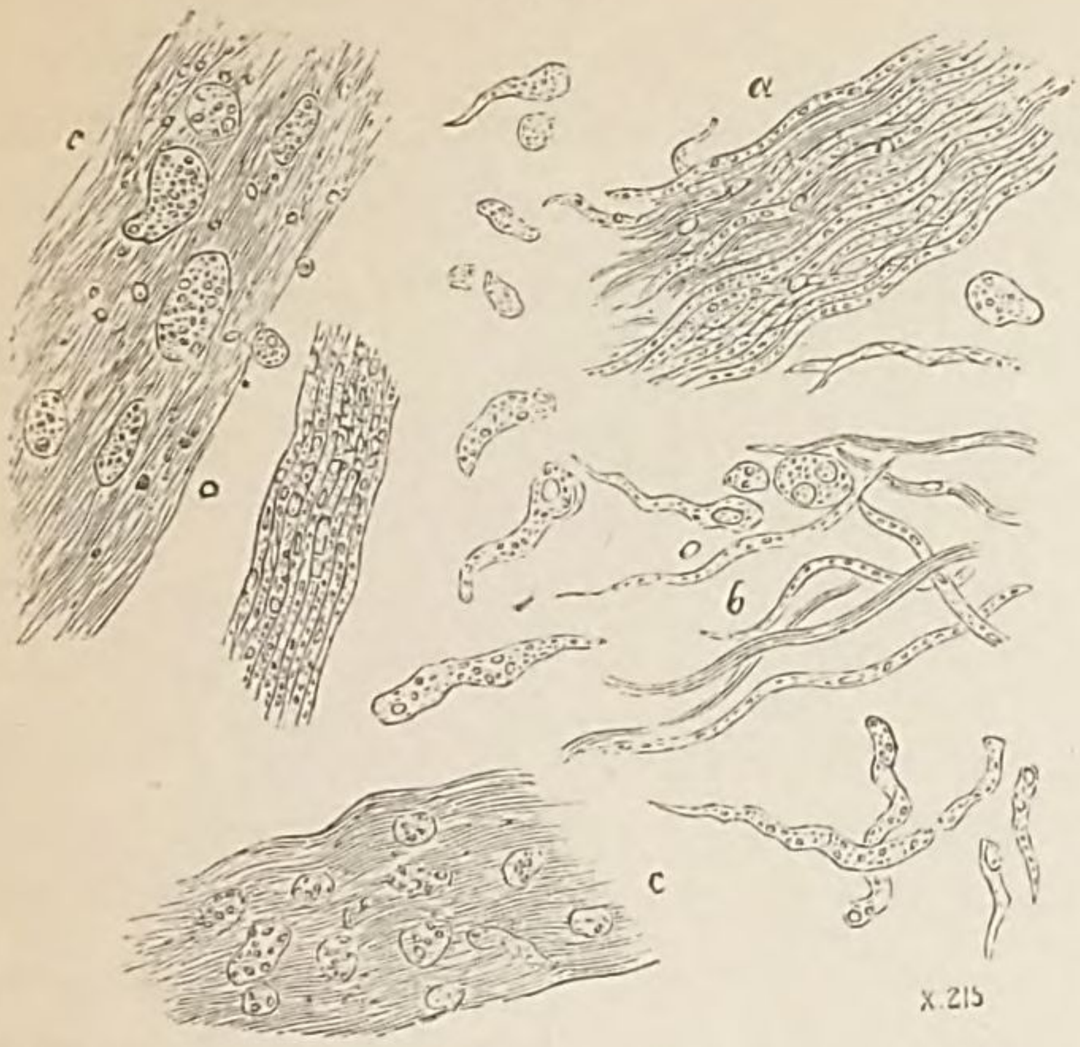
The walls of cysts are composed of fibrous tissue, and not unfrequently bone is deposited in them. They vary much in vascularity and sometimes the lining membrane is soft and spongy, occasionally covered with small papillary elevations and invested with a cellular layer. The character and number of the cells vary much.

The character of the fluid found in many cystic growths is described in page 270.

428. Cholesteatoma is a rare form of tumour, which was first described by Müller. Besides containing much fatty matter and crystalline plates of cholesterine, the soft, pulpy material of which these tumours consist, is composed of a number of glistening pearly scales, which may be easily separated into very thin laminæ. Upon examining these with a power of 200 diameters, they are seen to consist of egg-shaped vesicles. They are for the most part perfectly clear, but some exhibit a slightly granular appearance. Others again resemble cells of the epidermis which have been soaked in nitric acid. Of the precise mode of growth and origin of these peculiar cells I have been able to ascertain very little, but it is probable they are developed from modified cells of a sebaceous gland. The peculiar structure of these tumours is represented in pl. LXXII, fig. 3, the specimen having been sent me by Mr. Simon. The chemical composition of this tumour was as follows:—

* See "Archives of Medicine," No. I, page 33.

Fig. 1.

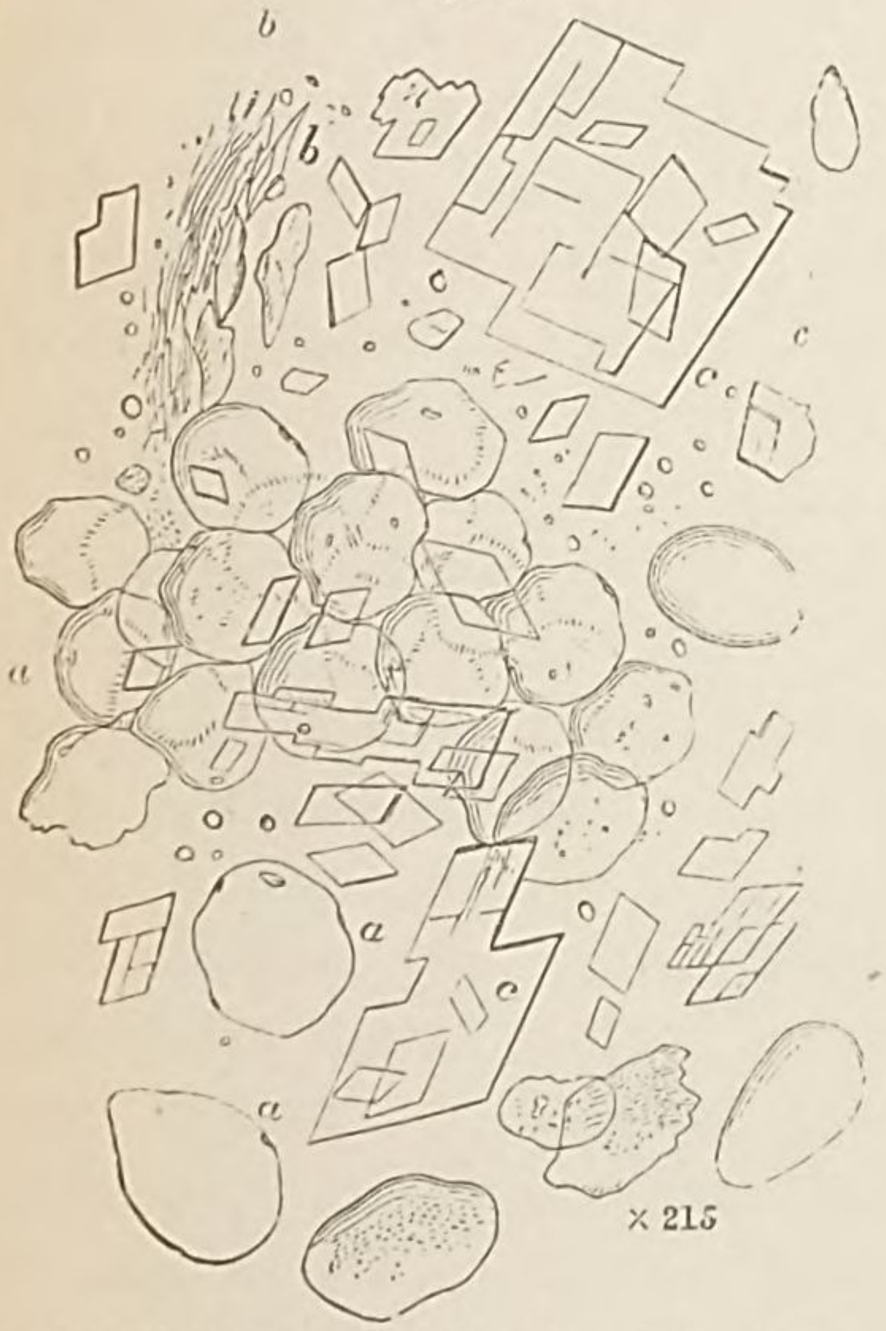


Fibroid tumour, from the testicle. × 215 *a*, *b*, fibres separated from each other. *c*, portion of the tumour in which the separate cells are not distinct. Collections of oil globules are scattered through this part of the mass. Fibre cells of various forms are represented p 460

Fig. 2.

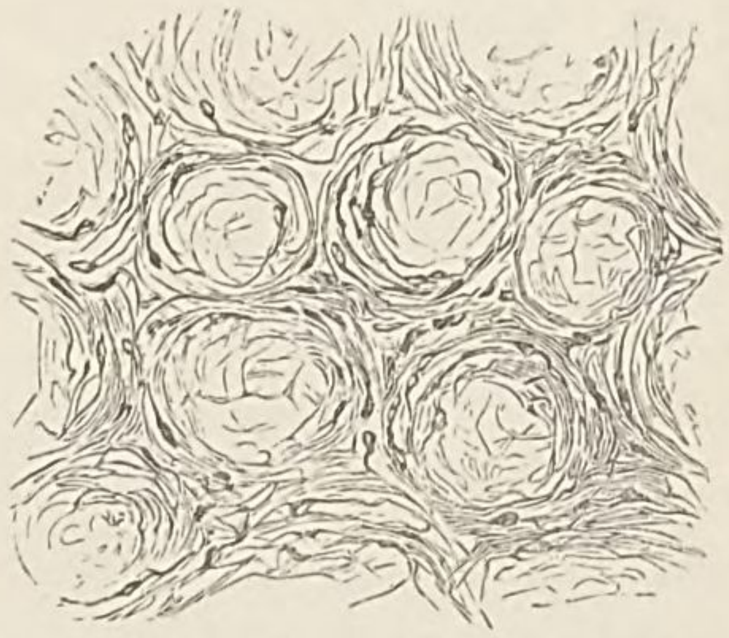
Softened cerebral tissue surrounding a cavity in the posterior part of the optic thalamus and the corpus striatum, and which extended some distance into the white substance of the hemisphere The cavity contained fluid and also a firm clot.

Fig. 3.



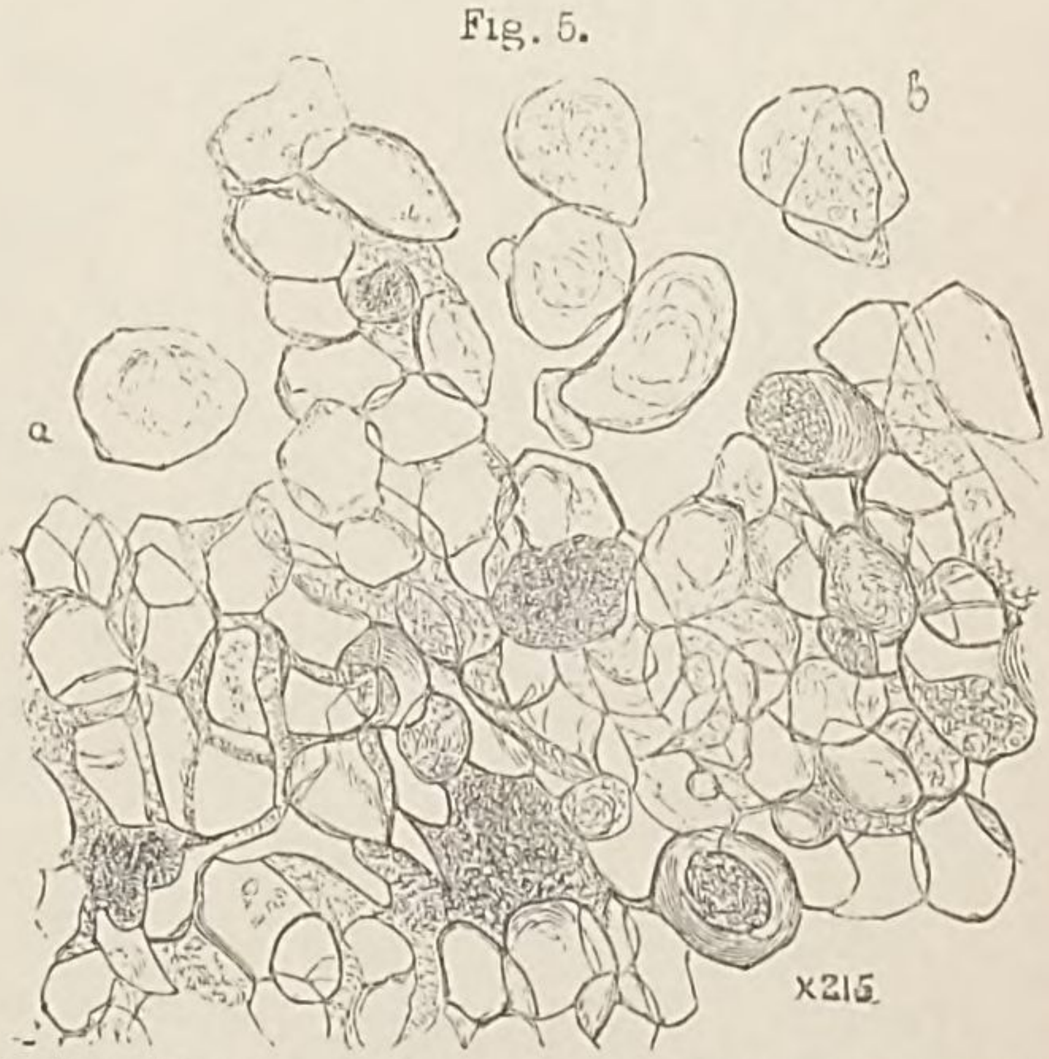
Cholesteatoma. *a*, large cells, of which the laminæ forming the pearly scales were composed; some of these are shrivelled and flattened, resembling the superficial scales of the epidermis. *b*, fibrous tissue from the inner surface of the capsule. *c*, crystals of cholesterine; oil globules and granular matter are seen in various parts of the field. p 461.

Fig. 4.



Mucous tissue from umbilical cord, showing its structure and mode of growth × 130. p. 461.

Fig. 5.



Section of tumour, situated under the tongue of a girl, aged 25. The dark shaded portions consist of fatty matter *a*, separate cell membrane. *b*, ditto, resembling cells of epithelium The structure of this tumour is closely allied to that of a cholesteatomatous tumour. p. 465.

1000th of an inch |___| × 215.

" " |_____| × 700. [To face page 464

		In 100 parts of solid matter.
Water	87·78	
Solid matter	12·22	
Extractive soluble in water and alcohol	3·119	25·52
Extractive soluble in water only ...	1·030	8·44
Fixed alkaline salts, consisting of sulphates, chlorides, phosphates, carbonates, with a trace of iron	·396	3·24
Fatty matter	·053	·43
Albuminous matter insoluble in boiling water	6·999	57·27
Earthy salts, consisting of phosphate and sulphate of lime ...	·608	4·97

The extractive matter soluble in alcohol had the same peculiar smell as the mass itself. The odorous material was volatile, and was present in the fluid which passed over in distillation in considerable quantity. The fatty matter was treated with alcohol, but no cholesterine crystallized out, probably in consequence of being protected from its action by the hard fat present. The total quantity of fatty matter was so small that no further experiments could be resorted to. It should be borne in mind, that an amount of cholesterine, which when examined in the microscope would be accounted considerable, is often so small as not to be appreciable by the balance.—"Archives of Medicine," No. I, page 42.

429. Rare form of Tumour beneath the Tongue of a Girl aged 25.—The tumour projected beneath the chin, and extended upwards into the mouth. It had been growing for about two years. It was opened by Sir W. Fergusson, and about an ounce and a half of a soft pultaceous mass was removed.

Analysis—1000 grains contained—

Water	838·72
Solid matter	161·28
Extractive matter	13·44
Alkaline salts...	·68
Fatty matter	45·00
Tissue, &c., insoluble	99·88
Earthy salts	2·28

The microscopical characters of the tumour were peculiar. These are represented in fig. 5, pl. LXXII. The mass was found to be composed of numerous cells, like adipose vesicles, filled with fatty matter, but some appeared nearly empty, and closely resembled cells of squamous epithelium, *a* and *b*. The microscopical characters of the

contents somewhat resembled those of cholesteatoma, but no plates of cholesterine were present in the specimen. In structure, however, this tumour was closely allied to the cholesteatomatous group. "Archives of Medicine," vol. i, p. 318.

430. Syphilitic Growths.—The structure of morbid growths which are due to the growth and multiplication of syphilitic bioplasm varies according to the particular tissues involved, the amount of the special morbid growth developed in one particular situation, and its age. In one case there may be merely what would be generally, but I think incorrectly, termed an "infiltration" of a membranous texture like the periosteum with minute bioplasts; in another a tumour may result and may grow as large as a walnut, consisting internally of a soft whitish matter and externally of a more or less condensed fibroid structure almost resembling a capsule.

At an early period of its development every syphilitic morbid growth is found to consist entirely of multitudes of minute bioplasts, which could not be distinguished from those developed in the course of other, or of ordinary inflammations. Years afterwards, however, nothing may be found except a slight accumulation or increase of ordinary connective tissue, or we may discover yellowish material not unlike caseous deposit—neither of which pathological formations are peculiar to syphilis.

In the liver syphilitic growths are not unfrequently found as small isolated tumours connected with the capsule, or formed here and there in the substance of the organ. It is probable that these commenced to grow in the portal canals. At the outer part of each separate mass there is usually a certain thickness of connective tissue, in the meshes of which a number of small bioplasts are to be demonstrated. The bulk of the tumour in its growing state is undoubtedly composed of roundish bioplasts. The oldest part of the mass is in the centre and it is here that the death of the bioplasm and its conversion into caseous-like matter first begins. As the tumour advances in age, a good deal of the débris resulting from the death of the living particles is absorbed and irregular contraction of the mass occurs. The tissue in the neighbourhood is often drawn or more or less puckered. Such are the broad characteristics which justify us in regarding certain morbid growths as syphilitic in their nature and origin.

The syphilitic "poison," or "virus," or "contagium" consists of a minute bioplast, which, like many other forms of morbid bioplasm when introduced into an organism whose nutrient fluids are in a favourable condition, grows and multiplies. The whole system may be infected and various local or general diseases may be the consequence. Such is the persistent tenacity of the bioplasm that it may live and grow for many years. Dormant perhaps for so long a time as to lead

us to believe it has been thoroughly destroyed and eradicated, but bursting into new and active growth when least expected and after its invasion had been forgotten. It is difficult to believe, but it is, nevertheless, an undoubted fact, that in too many instances ravages are produced in most important tissues in these cases by the growth and multiplication of germs that may have been quiescent for years. In this way most serious and fatal lesions may occur in important parts of the nerve centres, and there is hardly a tissue in the body that can be regarded as proof against the attack of the syphilitic bioplast.

From the resemblance of the syphilitic bioplasts to lymph corpuscles, syphilitic growths, like tubercles, have of course been pronounced to belong to the lymphatic category; but although no doubt in this class of pathological changes, as in many more, lymphatics and lymph corpuscles are involved, our knowledge does not justify us at present in regarding syphilis as a disease especially of the lymphatic system.

The syphilitic bioplast has been looked upon as a sporule, microzyme, or minute fungus germ, but it is not of this nature. We are familiar with many diseases of epithelium which are due to fungi, and which can be shown to be very different from affections of the same tissue dependent upon syphilis. Thus *Tinea versicolor* or *Chloasma* may be distinguished from roseoloid and other syphilitic eruptions by the presence of sporules of fungi in the altered epithelial cells. The syphilitic growth is due not to the presence of fungi or other bioplasts *in* the old epithelial cells, but to the changes resulting from the growth and multiplication of the syphilitic germ particles in and amongst the bioplasts at a time when they were in an early stage of growth.

It is certainly possible that syphilitic germ particles might invade individual elementary parts, as for example an epithelial cell or an elementary part of the liver, kidney, brain, or other organ. Although I am, of course, unable to bring forward actual proof of this notion, I have no doubt that important considerations may be advanced in its support. It is to be remarked that the morbid change invariably begins amongst the young and growing bioplasts of a texture, and it is quite conceivable that but only one may in the first instance be invaded by the syphilitic bioplast or germ particle. This last growing and multiplying amongst the particles of normal bioplasm soon causes derangement of the latter and the destruction of many. The neighbouring normal bioplasts necessarily suffer from the more vigorous growth of the morbid germ particles, and participate with these in experiencing an undue supply of nutrient material. Hence the "inflammation," the production of "inflammatory products," which invariably accompanies this and allied pathological processes. Growing and multiplying, the syphilitic germ particles may infect neighbouring normal elementary parts, and not a few syphilis germs of very minute

dimensions insinuating themselves between these cells may make their way to a distance or pass into lymphatics or blood vessels, and thus infect other tissues and distant parts. Such a doctrine is in accordance with many facts well known to those familiar with the practical side of the question. The advantage of the early use of caustics and other bioplasm killing remedies, the internal use of iodide of potassium, and indeed not a few circumstances which are inexplicable upon other hypotheses, may be reasonably accounted for.

The syphilitic bioplast or disease germ may live for a long time after it has entered the system if the conditions are not changed in such a way as to interfere with its growth and multiplication. The minute particles may make their way into every tissue, and by their growth and multiplication, or by the products resulting from their degeneration, decay, and death, seriously interfere with the discharge of function, and render impossible the formation of tissue which possesses the ordinary firmness and powers of resistance manifested by the healthy texture. There is not a tissue or organ of the body that may not be invaded and damaged by syphilitic disease; but there are certain textures which are more prone to suffer than others, and many of these are also remarkable as being tissues which are the seat of pathological changes which occur in "scrofulous persons," and those who inherit a tendency to tubercular diseases. But the syphilitic bioplast is, upon the whole, more virulent and more tenacious of life than other forms of morbid bioplasm, and its peculiar tendencies to damage various nervous organs in middle life and to destroy nerve-fibres and nerve-cells in the central nervous system are but too remarkable and too frequently observed.

431. Epithelial Growths.—*Epithelial Cancer.*—*Epithelioma.*—These tumours resemble the cancerous growths more closely than any other structures. The distinctive characters have been investigated by Paget, Bennett, Lebert, Walshe, and many more.

The following forms of disease belong to this category:—*Cancer of the lip*, *noli me tangere*, *cauliflower excrescence of the uterus*, *chimney sweeps' cancer*, and some others. *Warts* consist merely of a superabundant formation and accumulation of the epithelial cells of the cuticle; and tubercles, which occur on the external genital organs, have a very similar structure. These are both very different from epithelial cancer, and grow more slowly. As cancer of the lip, tongue, &c., advances in growth, fissures are formed, in which an abundant growth of epithelium takes place, accompanied with an ichorous discharge. The papillæ have become much modified, and, like other textures, entering into the formation of the so-called cancerous tissue, grow very irregularly. As the disease gradually advances, it invades deeper structures.

If a thin section of a well-developed epithelial growth be examined, interspaces will be observed, pl. LXXIII, fig. 1, upon the walls of which

the cells are found, and in some forms appear to grow. The cells often seemed to be arranged in laminæ; they do not vary so much in size and form as the cells of true cancer; the masses of bioplasm "nuclei," do not differ so much in size; they rarely contain large nucleoli, and usually adhere to each other by their margins, pl. LXXIII, fig. 2; frequently three or four, or more, will be found united together. In fact, these cells resemble, in their general characters, the ordinary epithelial cells of the surface upon which the growth is developed, but they are softer and contain more water, and grow much more quickly.

As the growth of the tumour advances, the cells become larger and softer. They grow more quickly, lose all resemblance to epithelial cells, and contain a large number of bioplasts and bioplasm particles.

Many of these so-called *epithelial growths* may become true *cancers*. They commence in connection with some epithelial tissue, as the cuticle of skin, or of mucous membrane, or in the duct or follicles of a gland.

The chief differences said to exist between epithelial cancers and well marked cancerous growths may be tabulated as follows :—

Cancerous.	*Cancer-like Growths.*
Cells not connected with the matrix in a regular manner, or forming laminæ.	Cells connected with the matrix, often forming distinct laminæ.
Cells differing much from each other in size and form.	Cells resembling each other in size and general outline.
Cells readily separable from each other.	Cells often cohering by their edges, which generally form straight lines; three or four cells being frequently found united together.
Cells not connected together at their margins; their edges seldom forming straight lines.	
Cells containing several smaller cells in their interior often met with.	Cells usually containing one nucleus.
Nuclei varying much in size and number in different cells.	Nuclei not varying much in size in different cells.
Juice scraped from the cut surface containing many cells floating freely in the fluid, and not connected with each other.	Juice scraped from the cut surface, containing small collections of cells, which are often connected with each other.

A beautiful specimen of cells from a cancerous growth, originating probably in the epithelium of the surface, or of a gland from the pharynx, is represented in pl. LXXIV. The cells were expectorated by the patient during life. I am indebted to my friend, Mr. Newham, of Bury St. Edmunds, for the specimen and account of the case published in the fifth number of the "Archives of Medicine."

It will be observed that the cells represented differ much from one another, and are not all composed of the same material. Some refract light differently to others, as indicated by the different varieties of shading, and there is an absence of that granular appearance which is observed in the greater number of specimens figured. The cellular appearance of many of the bodies in question is fallacious, and many that would be termed "mother cells" are only masses of soft formed material with nuclei (bioplasm centres) irregularly scattered through

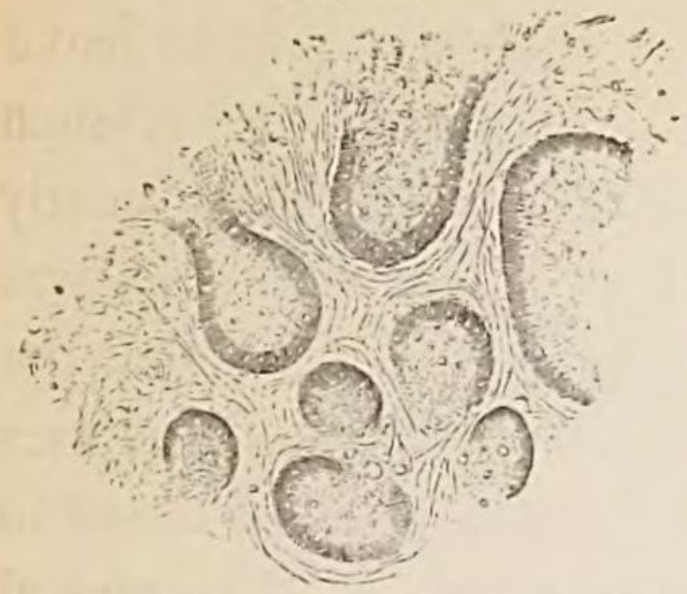
them. In some instances these have broken in such a way as to leave cavities into which the bioplasm evidently fitted. At *p*, fig. 1, such a mass is seen, and at the lower portion is a cell-like piece nearly detached, with others which are quite separated. The specimen was not treated with any reagent. Water was not even added, so that the appearances represented are not produced by any artificial processes whatever. A portion of the mass removed after death is represented in fig. 2, and in fig. 3, the microscopical characters of one of the cervical glands are indicated. Other forms of cancer are represented in pl. LXXIII.

432. Cancer.—A cancerous growth may be described as consisting of a fibrous matrix, pl. LXXIII, fig. 4, more or less abundant, and arranged so as to form areolæ, or interspaces, *a*, upon the walls of which the vessels ramify. These interspaces contain cells in considerable number, suspended in a more or less viscid fluid, with much granular matter which is found when examined in a perfectly recent state under very high powers, to consist of *a*, minute particles of living bioplasm, the germs from which new cells may be formed; *b*, fatty matter as minute granules or molecules, and globules, and minute crystals of fatty matter; and *c*, shreds of fibrous tissue.

The great difficulty of deciding as to the cancerous or non-cancerous nature of a tumour, arises principally from the fact, that no single element of which the structure is composed, can be looked upon as characteristic of the cancerous form of growth only. Neither the character of the cells, nor the structure of the matrix, nor the arrangement of the elementary constituents can separately determine the point, and it is only by carefully noting the collective appearances observed upon microscopical examination, that we can decide. In the great majority of cases, however, it is possible to speak with tolerable certainty; but at the same time it must be borne in mind that instances come under notice from time to time, in which the most careful and experienced observers would be unable, from a microscopical examination alone, to decide positively as to the nature of the tumour. A well-defined cancerous growth, however, in its microscopical characters, does not resemble, and cannot be confounded with, any healthy texture; while many of the non-malignant tumours, in their essential characters, bear great similarity to certain healthy tissues, or are actually identical with them in structure.

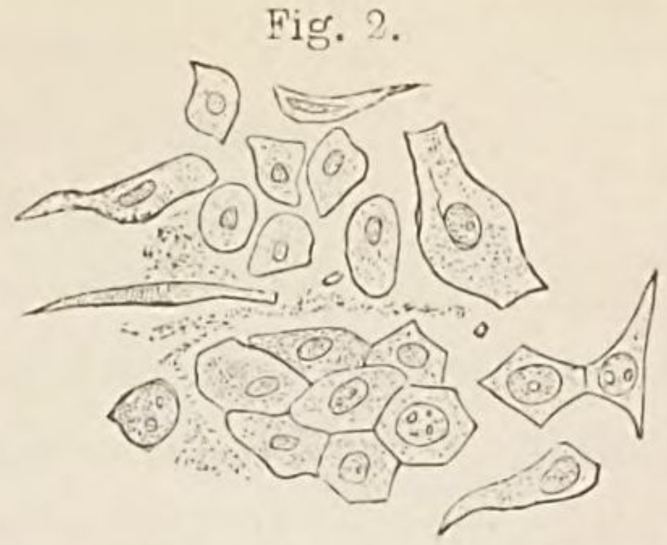
From the freshly cut surface of a *cancerous tumour*, a more or less turbid juice exudes, which, upon examination in the microscope, is found to contain cells varying much in size and form, as well as in the character of their contents; a few fragments of fibrous tissue; a number of free oil globules, and, perhaps, a few cells containing oil globules; and much free granular matter, pl. LXXIII, fig. 4. Upon examining a thin section made with a Valentin's knife, the relation of these

Fig. 1.



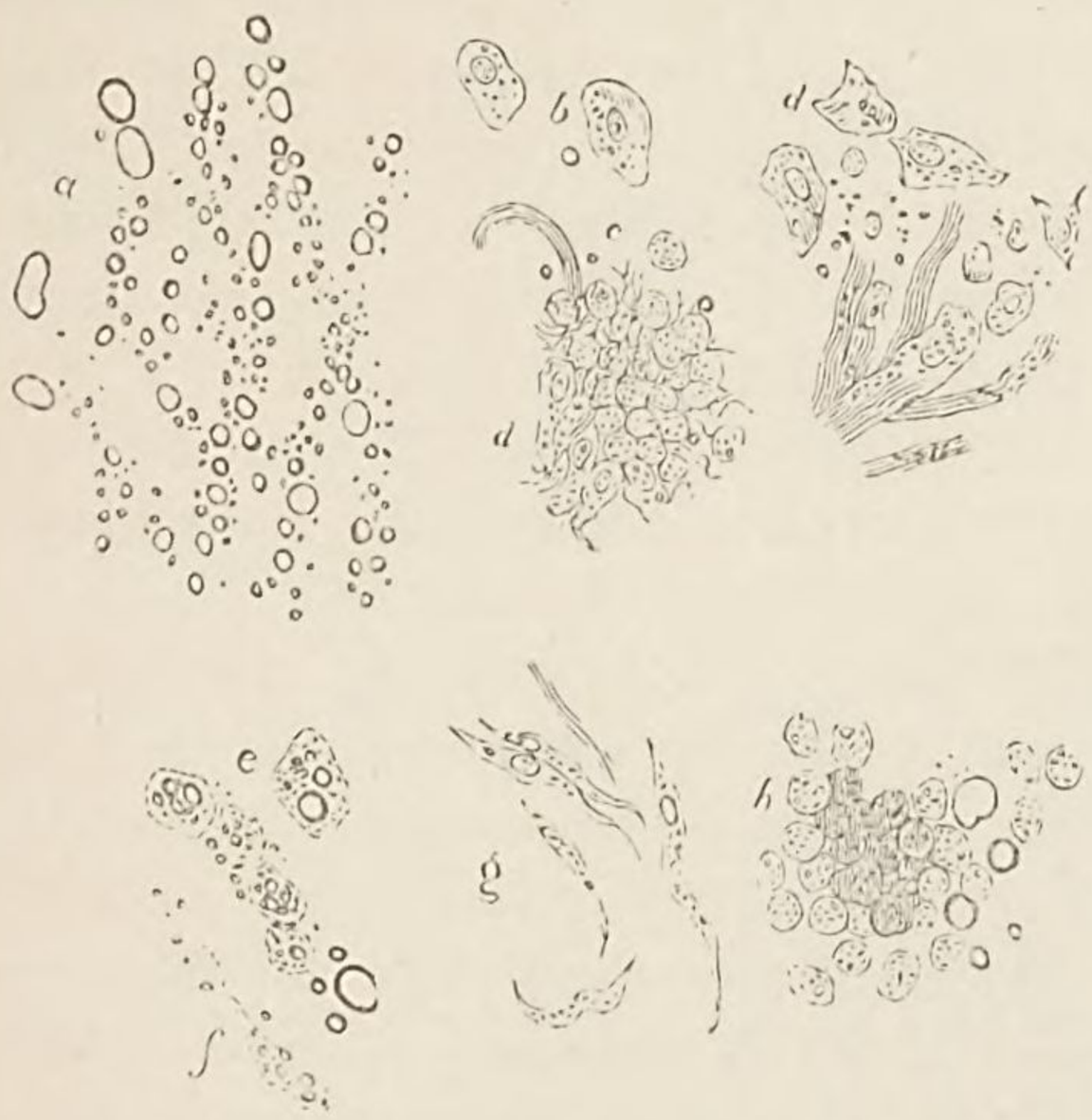
Epithelial cancer, showing the stroma and the situation of the cells. × 42. p. 468.

Fig. 2.



Elementary parts resembling ordinary epithelium, from an epithelial cancer × 215. p. 468.

Fig. 3.



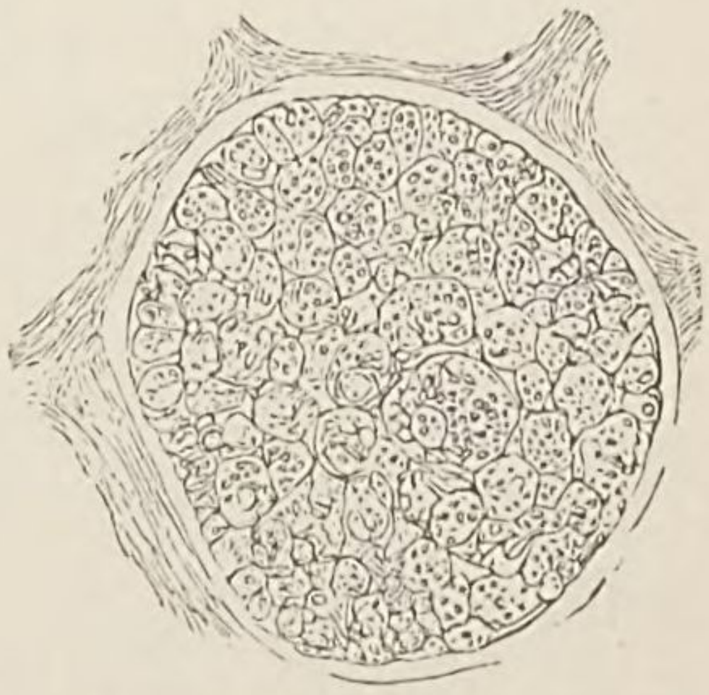
Drawings from a case of cancer diffused through the entire liver. Sent by Mr. Robert Ceely, of Aylesbury, Case reported in 'Archives,' vol. I., p 129

The specimens were taken from the collections of cancerous matter, and from the intervals between them. *a*, remains of the secreting structure of the lobules *b*, liver cells from the interval between two collections. *c*, *d*, also from the surface of one of the cancerous masses, showing delicate fibrous tissue, cells, and fibre cells. *e*, remains of secreting cells in the interval between the cancerous collections. *g*, fibre cells from the surface of a collection. *h*, cells from a short distance within one of the tumours. × 215. p 470.

Fig. 4.

From a cancerous tumour. *a*, portions of fibrous matrix. *b*, bioplasts, many of them containing smaller ones in the interior. × 215 p. 469.

Fig. 5.



Small cancerous tumour from a portal canal, containing a number of bioplasts. × 215 diameters. From a woman, aged fifty-one, who died of medullary cancer of the liver, in King's College Hospital. M B., 572. p 471.

1000th of an inch ⌴ × 42.

" " ⌴ × 215.

[To face page 470.

structural elements to each other may be observed. The fibres will be seen to form meshes or interspaces, in which the cells and fluid are contained, fig. 4 *a*. In some instances the fibres resemble those of ordinary areolar tissue; sometimes they consist chiefly of fibres resembling those of yellow elastic tissue; and not unfrequently the fibres become perfectly transparent upon being treated with acetic acid, showing the absence of the yellow element. Amongst these fibres, helping to form the boundaries of the spaces, are the capillary vessels which cannot be discerned in many cases unless they are injected. Many forms of cancer prepared according to the directions given in Chapter VII, yield most beautiful and instructive microscopical specimens.

The cells of cancerous tumours vary much in size and form: they may be perfectly round, or prolonged at either end into delicate fibres, or of most irregular outline, pl. LXXIII, figs. 1, 2, 3, 4, 5, pl. LXXIV. They contain at least one large mass of growing bioplasm, "nucleus," but very often two are met with, and not unfrequently many more may be observed. These are formed by the subdivision of the centre. The masses of bioplasm "nuclei" of different cancer cells often differ much in size. They generally contain several granules, and much granular matter exists between them and the formed material or "cell wall." Cells are often found which contain several smaller cells in their interior, and have, on this account, been termed "mother-cells," fig. 4, pl. LXXIII, fig. 1, *b*, *d*, *q*, etc., pl. LXXIV. The cells readily separate from one another, and exhibit no tendency to aggregate together, nor do they appear ever to have been adherent to each other at their margins. Fig. 5, pl. LXXIII, is a beautiful example of a very young growing cancerous tumour, consisting of only a few cells which are multiplying rapidly.

The characters of many cancerous growths depend upon the locality in which they grow, and a cancer may assume the form of a solid, hard, or soft circumscribed tumour, a soft spongy mass, prone to spread in all directions, a highly vascular papillary growth, or other forms too numerous to mention. Cancerous growths also differ in density, colour, rapidity of growth, as well as in the form and character of the cells of which they are composed. It is impossible to lay down any definite characters which shall in every case serve to distinguish a cancerous tumour from other forms of morbid growths; but a tumour that has grown quickly, and from the cut surface of which a milky juice is poured out, which, upon microscopical examination, is found to consist principally of cells exhibiting the general characters above referred to, and arranged in the meshes of a fibrous stroma, may be pronounced to be of a cancerous nature.

True cancers are also supposed to originate invariably in epithelium, but no one has yet offered a plausible explanation of the early phenomena, which upon this hypothesis must be manifested in the indi-

vidual cell or cells which represent the first departure from the normal condition. What is the difference between the cell which, to use the favourite but highly-objectionable term, "proliferates," towards the production of mere inflammatory products, and that which is to be the parent of cancerous progeny? To me it seems far more probable that the cause of the cancer development operates at a period of time separated perhaps by many, many years from the period of the actual production of the cancer cells. Just as any complex tissue or organ was, as it were, laid down and prepared for long before its actual development is observable, so I believe it will be proved that those changes which determine the production of a cancer, say at the age of fifty, were initiated at an early age, and in many instances, even during embryonic life. And, as I have ventured to state already in the introduction to this chapter (page 453), I regard every kind of cancer as originating in the bioplasm which exists as *embryonic bioplasm*, even in the tissues of the adult. The actual changes characterizing the first departure from the normal state manifested by the bioplasm can only be presented to the imagination, and yet there are phenomena which precede these, of which the initial changes are a consequence; but it may be safely predicated that the development of the cancer is due to an irregularity in the occurrence of ordinary phenomena, to a sort of dislocation or derangement of the matter of the bioplasm rather than to the development of a new power or a new and forced combination of the material particles which constitute the bioplasm from which the cancer bioplasts proceed.

433. Melanoid Tumours.—The terms *melanosis*, *melanoma*, and *melanoid*, have been applied to those cellular tumours which contain a considerable quantity of pigment. The colour may vary from a darkish yellow to a purple or black, and the material of which the growth is composed consists of minute granules or small masses, varying much in shape and size. These are composed entirely of organic matter with a mere trace of iron, and are precipitated amongst the bioplasm at an early period of the growth of the cell, and they are therefore found in the substance of the cells in their fully formed state. The cells of many cancers, epithelial and others, contain much pigment, and these are consequently said to be *melanotic*.

The lungs and bronchial tubes of colliers, sweeps, and those who work in many factories, often contain a quantity of black material which has been introduced into the lungs suspended in the air breathed. This state has been termed *false melanosis*.

434. Examination of Morbid Growths.—In the first place the fluid or juice, if any exists on the free surface of the tumour, should be examined: secondly, the microscopical characters of the juice, which exudes from the freshly-cut surface should be ascertained: and, lastly,

Fig. 1.

Various forms of the so-called cancer 'cells' in sputum Many of these bodies are clearly not 'cells,' but fragments of a mass containing nuclei, or small masses of bioplasm irregularly scattered through it. At *p* is represented a piece from the lower part of which the nuclei and portions of the mass surrounding them have been broken off, leaving cup-shaped cavities in which they were lodged. p. 469.

Fig. 2.

Portion of the tumour itself, removed after death. p. 469.

Fig. 3.

Portion of a cervical gland. p. 469.

a thin section of the structure ought to be made, in order to determine the relation of the constituents of the tumour to each other, and especially the proportions in which the different elements are present. Its connection with surrounding structures may be ascertained by examining a thin section, which should include a portion of the adjacent texture; and these observations should be made first with low powers, and afterwards with a power of from 700 to 1800 diameters.

The disposition, arrangement, and general direction of the fibres in the fibrous portion of the tumour should be carefully noted, and the form, size, shape, and contents of the "cells," especially with reference to the presence or absence of oil globules or molecules, together with nuclei, or new centres one within the other in the bioplasts, should be especially dwelt upon. Every opportunity should be taken of carefully delineating the appearances observed, in order that the structure of one tumour may be compared with that of others, which may subsequently come under the observer's notice, and if the growth presents anything unusual, a section ought to be put up in some preservative fluid. It is extremely important that every opportunity of obtaining very minute cancerous tumours should be taken advantage of. More is to be learned with reference to the history and mode of growth of cancers, from microscopic tumours, than from those of large size. Accurate notes should be made of every examination, and these, with sketches, should be entered in a note book kept for the purpose.

The observer should examine tumours in several different parts. Sections may be made in different directions with Valentin's knife, an ordinary scalpel, or a strong knife, according to the consistence of the tumour. In examining the cells it is better to employ a little serum or gum-water, for sometimes if water alone be used, they become swollen and much altered.

The influence of certain chemical reagents upon the sections and portions scraped from the cut surface of the fresh tumour must be ascertained. The most important reagents in the examination of morbid growths are, acetic acid, solution of soda, and ether; but the stronger acids and other tests will occasionally be required. The two former are of advantage in rendering the matrix more transparent, and displaying the nuclei. Ether is sometimes required to ascertain if certain globules which resemble fatty matter, are really of this nature.

The method of preparation described in Chapter VII is eminently adapted for the investigation of morbid growths. Tumours of the most delicate structure may be injected by the process there described, and the bioplasm of the cells may be stained, and thus very beautiful and highly demonstrative specimens may be prepared. All may be preserved in strong glycerine or in the mixture of gelatine and glycerine. See page 53.

CHAPTER XX.

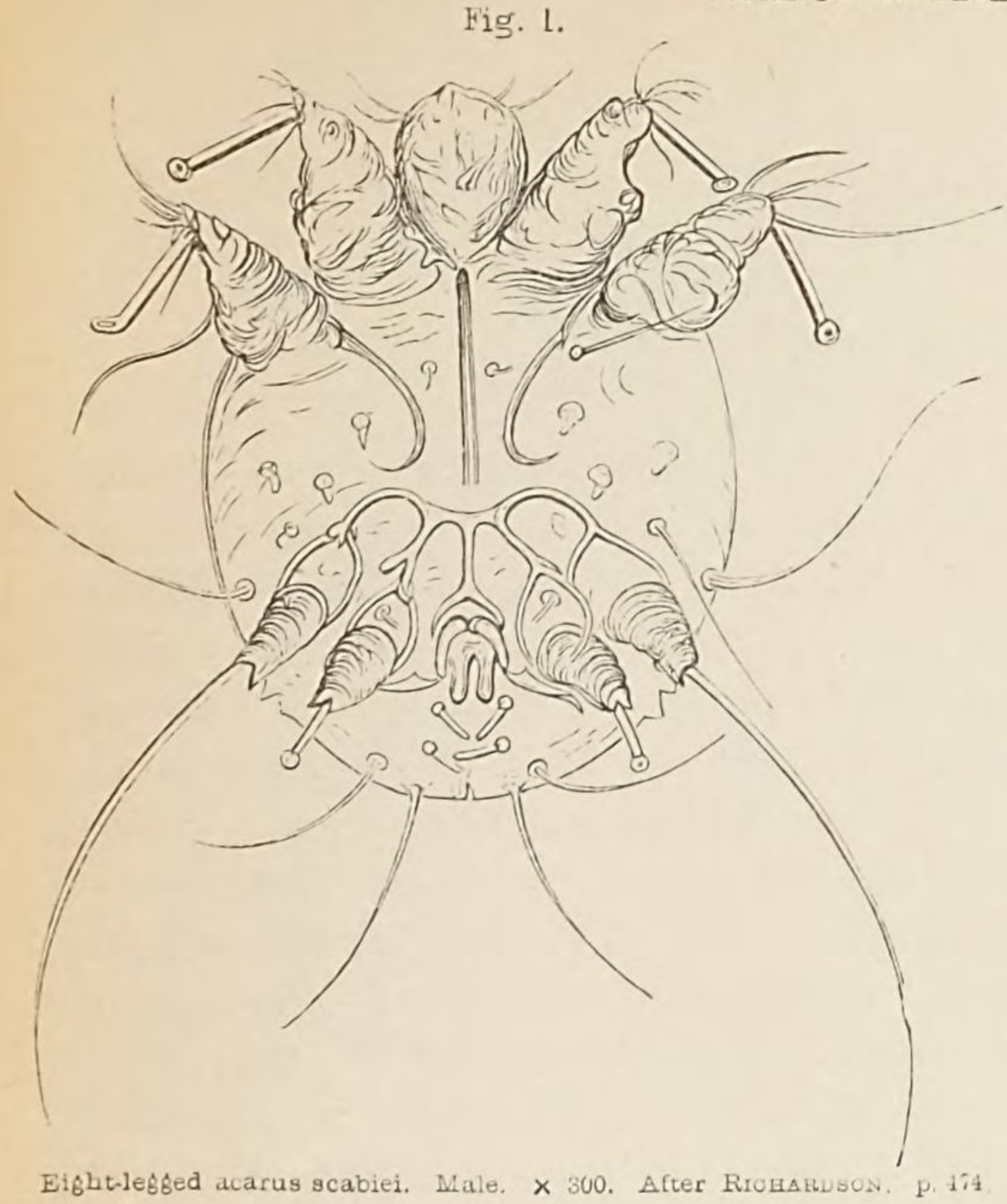
ANIMAL AND VEGETABLE PARASITES.—ANIMAL PARASITES.—*Acarus Scabiei.*—*Entozoon folliculorum*, or *Demodex.*—*Examination of Entozoa.*—*Tænia Solium*, *Tænia Mediocanellata.*—*Bothriocephalus Latus.*—*Hydatids*, *Echinococci.*—*Trichina Spiralis.*—*Filaria or Hæmatozoon of human blood.*—*Bilharzia Hæmatobium.*—*Other Entozoa.*—*Strongylus Gigas.*—PROTOZOA.—VEGETABLE PARASITIC STRUCTURES.—*Sarcina Ventriculi*, or *Merismopædia Ventriculi.*—*Other Forms of Algæ.*—*Leptothrix Buccalis.*—*Penicillium Glaucum.*—*Achorion Schœnleinii.*—*Psorosperms and Gregariniform bodies on hair.*—*Tricophyton Tonsurans.*—*Tricophyton Sporuloides.*—*Microsporon Mentagraphytes.*—*Microsporon Audouini.*—*Microsporon Furfur.*—*Puccinia Favi.*—*Chionyphe Carteri.*—*Early Stages of Fungi.*—*Oïdium.*—*Aphthæ*, *Muguet.*—*Diphtheria.*—*Fungus from External Meatus of the Ear.*—*Other forms of Fungi.*—*Of the Manner in which Fungi enter the System.*—*Examination of Vegetable Growths.*

IN this chapter I shall only refer to the structure and mode of examining some of the parasites which are of greatest interest to the microscopical observer. Of the larger and most widely distributed epozoa it is not necessary to speak in this work, and for a complete account of the entozoa which live upon the fluids of man and animals, I must refer the reader to the following well-known treatises: Cobbold on Entozoa—Die Menschlichen Parasiten von Rudolph Leuckart—Siebold, art. Parasiten in H. W. B. für Physiol. Th. II.—Küchenmeister über Menschl. Parasiten, translated for the Sydenham Society, and to others which will be found enumerated at the end of the present chapter.

ANIMAL PARASITES.

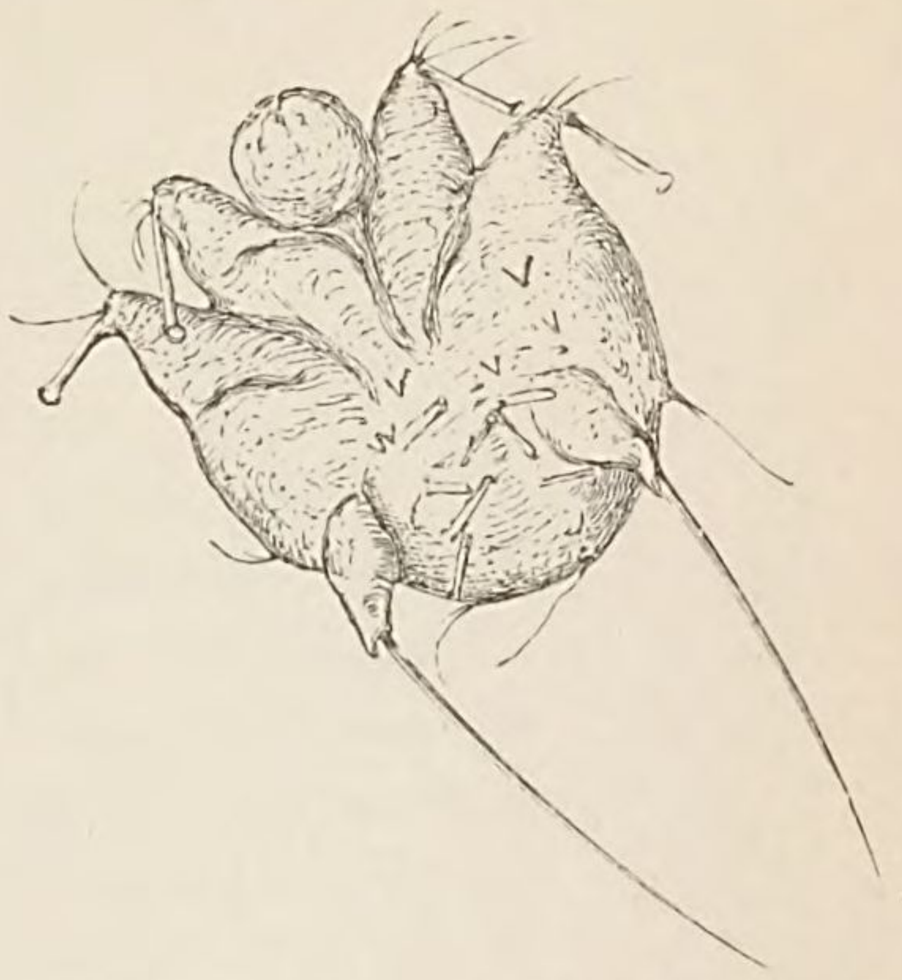
435. Acarus Scabiei, or Sarcoptes Hominis.—The itch acari are rather difficult to procure from the very mild cases of itch usually met with in this country. They may sometimes be extracted from the itch pustules or vesicles by passing a fine needle into the burrow, the opening of which is always at the side, and may be known by the presence of a little dark point. The male is very much smaller than the female. See figs. 1, 2, pl. LXXV, for which I am indebted to my friend Mr. Richardson, of Dublin. The difference of magnifying powers used

Fig. 1.



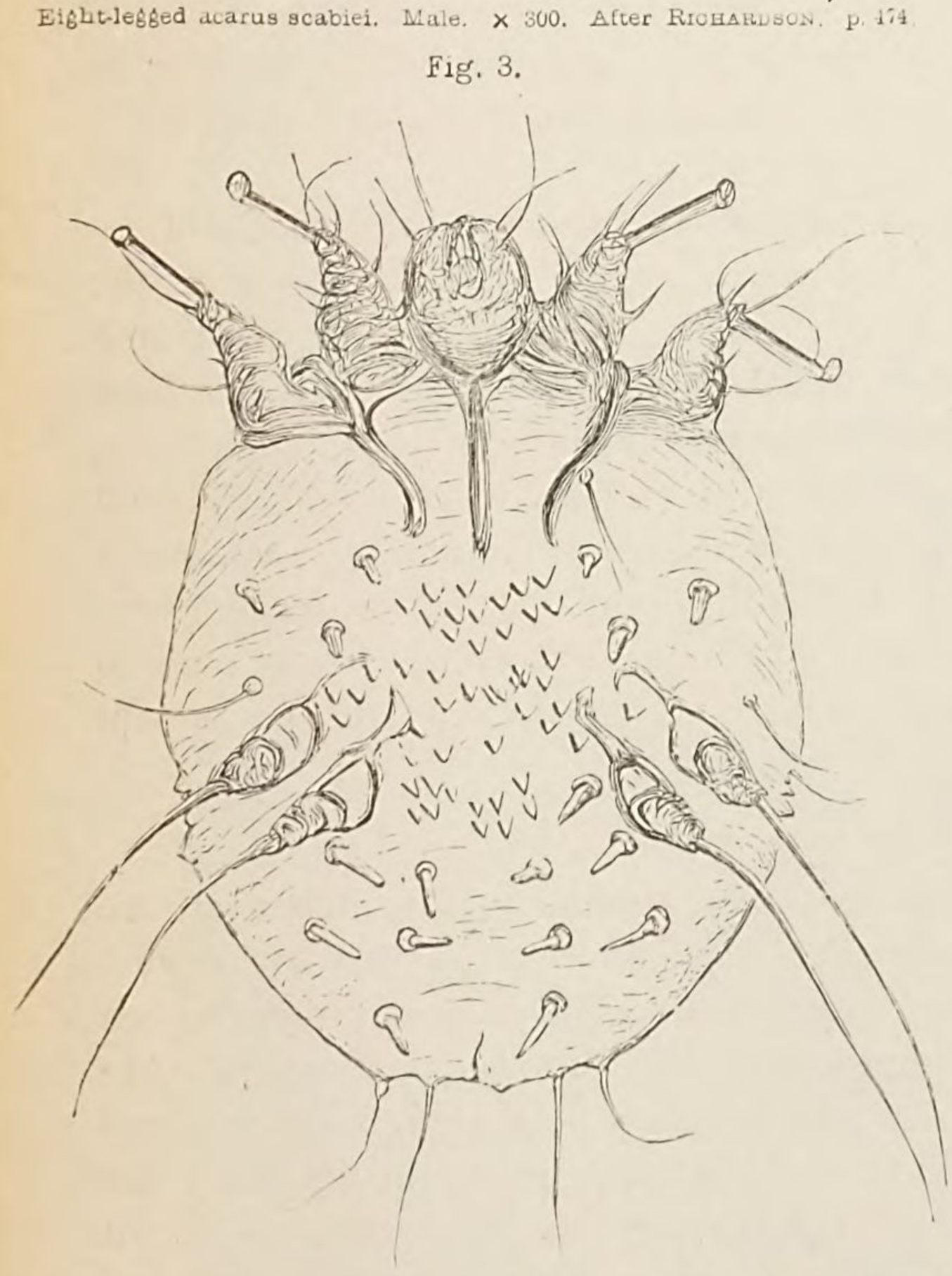
Eight-legged acarus scabiei. Male. × 300. After RICHARDSON. p. 474.

Fig. 2.



Six-legged acarus scabiei. × 200. After RICHARDSON. p 475.

Fig. 3.

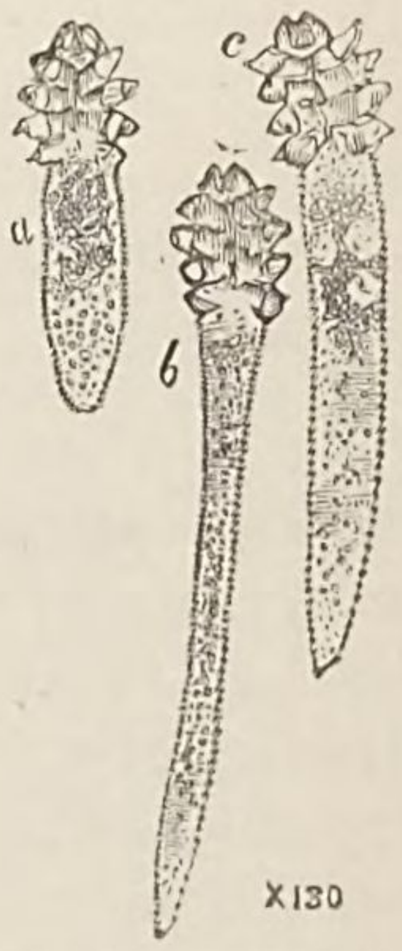


Eight legged acarus scabiei. Female. × 130. After RICHARDSON. p 474.

Fig. 4.

Eggs of acarus scabiei in various stages of development. After KUCHENMEISTER. p 475.

Fig. 5.



Entozoa folliculorum From the external auditory meatus. *a c*, short variety. *b*, long variety. × 130. p. 475.

1000th of an inch ⎽⎽ × 130.
" " ⎽⎽⎽⎽ × 215.
" " ⎽⎽⎽⎽⎽ × 300.

[To face page 474

in delineating the specimens must be noticed. The acari may be dried carefully at a gentle heat, and preserved in Canada balsam.

The itch mite bores into the skin from the outer surface. At first the direction of the gallery is perpendicular, but it passes obliquely through the cutis. In order to obtain the mite, the Corsican women pass a needle or other sharp pointed instrument into the opening in such a manner that it might be forced below the mite, the portion of skin elevated, and the acarus turned out. The process requires some practice and dexterity for its performance. A sharply pointed thin knife is an efficient instrument for obtaining the acarus. In order to examine the galleries the creature makes, the skin, with the vesicle or pustule, is to be pinched up, and the latter shorn off with a knife or scissors. It is then inverted and examined in a fluid or in glycerine, or allowed to dry slowly on a glass slide, when it may be mounted in Canada balsam.

In bad cases of itch, *crusts* may be removed which contain numerous specimens of the acarus in various stages of development with ova. Mr. B. W. Richardson, of Dublin, received from Dr. Neligan a piece of scab from a *case of Norway itch*, which, although not more than a quarter of an inch square, contained one hundred specimens in it. They were all *six legged*, as represented in pl. LXXV, fig. 2. In a case which occurred at Wurzburg, and is alluded to by Mr. Anderson, a piece of the crust "not half a line square, contained two females, eight six-legged young, and twenty-one pieces of acari, six eggs, and fifty-three egg shells. In the deepest and softest parts of the crusts, masses of living acari wallowed and tumbled about."*

The hexapod acari are not, as some have supposed, a distinct species, but merely young specimens of the ordinary acarus which have not moulted. They are found in numbers never met with in this country in the severe forms of Norway itch. The female lays more than fifty eggs. Ova of the acarus scabiei, in different stages of development, are represented in fig. 4, which have been copied from Küchenmeister.

436. The Entozoon Folliculorum is generally present in the follicles of the skin of the scalp, nose, chin, and other parts of the face. It may usually be procured very readily from the nose, by squeezing out the contents of the sebaceous follicles by pressing the skin firmly between the finger and thumb, or between two of the finger nails. The white cheesy matter thus squeezed out must be torn with needles, and then placed on a slide in a drop of oil, and covered with thin glass. One or two of the entozoa will usually be found. There are two varieties, and these are constantly met with in the same individual. One is much

* Mr. B. W. Richardson, "On hexapod acari scabiei, from Norway itch." Dublin Medical Press, June 7, 1865.

longer, and the body more thin and taper than the other, fig. 5, *a*, *b*, *c*, pl. LXXV.

I have found them in considerable number in the wax which collects in the ear. If the wax is tolerably moist the addition of oil is unnecessary.

Small parasites may be stained with carmine and preserved in strong glycerine. After proper soaking in glycerine they may be preserved in the glycerine jelly, or they may be carefully dried and mounted in balsam. Potash and soda dissolve the soft parts, and thus the skin may be made beautifully displayed.

437. Tape Worms.—*Tænia Solium.*—The common English tape worm is often met with. A fresh joint may be placed under the microscope and examined with low powers. If dried upon a glass slide, and mounted in Canada balsam, it makes a very instructive preparation. The ovaries of many joints are often distended with ova some of which should be squeezed out and mounted separately. See pl. LXXVIII, page 484.

The mode of development of the tape worm is now quite understood, and it has been proved experimentally that the cysticercus cellulosæ becomes in the stomach transformed into Tænia solium. The cysticercus is introduced into the organism in measly pork; it has been remarked that cases of tape worm are most common in those parts where much pork is eaten. The cysticercus cellulosæ has been met with in several cases in the human eye, as in the anterior chamber, in the vitreous, and in some instances it has migrated into the brain.

Tænia Mediocanellata. This tape worm is developed from a cysticercus infesting cattle, and derives its name from the peculiar distribution of the water vessels in the head. It is larger every way than tænia solium. The head is very large, flattened, and hookless, with four suckers much larger than those of tænia solium, surrounded by much black pigment. Dr. Cobbold remarks that this hookless tape worm is as common in this country as the tænia solium. In pl. LXXVI, fig. 1, is a drawing of a beautiful specimen of the head of this entozoon which was passed by one of my patients in the hospital after oil of male fern.

Bothriocephalus latus is the tape worm most common in Russia, Sweden, Poland, and Switzerland. It is seldom met with in this country, and out of upwards of 100 cases of tape worm I have only obtained one specimen of the bothriocephalus. The head is elongated and destitute of hooks and suckers. This parasite may be examined and preserved in the same manner as the common tape worm.

438. Means of Procuring the Head of the Tape Worm.—It may be advantageous just to refer to the most effectual manner of obtaining the head of the tape worm. Of all the remedies I have seen tried, the etherial oil of male fern is certainly the most efficacious. Out of about

thirty cases which I carefully watched in 1851, when I was house physician to King's College Hospital, the head was expelled in six or seven. Some of the patients had been treated with kousso, and others with the oil of male fern. All the successful cases had been treated with the latter; indeed, although I have seen many cases treated with kousso, I never was successful in finding the head; the greater part of the worm, however, was invariably expelled. The oil of male fern is to be administered as follows;—two drachms to half an ounce, according to the age and strength of the patient, are suspended in eight ounces of water, with the aid of mucilage. After fasting for twenty-four hours (only a little water, or, at most, milk being allowed), the patient is made to take the draught early in the morning, and an hour or an hour and a half afterwards, a dose of castor oil is to be given. The worm is usually expelled in the course of the day. The fasting appears to be a very important part of the treatment, and it seems essential that the oil should be suspended in a large quantity of water. I have since obtained many entire worms in this manner.

The head may be examined in fluid with an inch object-glass as an opaque preparation, or it may be put up in balsam, but it must be dried with great care. I have found that specimens of tape worm may be preserved exceedingly well in strong glycerine to which a little acetic acid has been added.

439. Hydatids. Echinococci.—Hydatids may occasionally be obtained from bodies in the post mortem theatre. They are usually found in large cysts, occupying a considerable portion of the liver. The parent cyst is often surrounded by a layer of purulent fluid. Upon opening this parent cyst numerous smaller round cysts (*acephalocysts*) with much fluid, escape. The walls of the cysts are usually quite white, not unlike the boiled white of egg; and they vary much in thickness. The thick, white membrane consists of several superimposed laminæ which increase in number as the cyst advances in age, the new layers being deposited from within. See fig. 7, pl. LXXVI. These may be well seen in a thin section of the walls of the cyst. Often a considerable number of crystals of triple phosphate will be found, especially if the hydatid be not quite fresh. The structure of the wall appears homogeneous or at most slightly granular, as if it had been merely deposited by the inner germinal membrane from the inner surface of which the echinococci and new cysts are formed.*

The granular appearance of the inner membrane arises from the presence of little elevations with which the surface is studded. By scraping these gently with a knife, not unfrequently many echinococci will be removed. The echinococci may also be obtained by allowing

* See also a communication by Dr. Hyde Salter, in the fifth volume of the "Transactions of the Pathological Society," page 303.

the fluid contents of the acephalocysts to flow into a conical glass. After a short time the echinococci sink, and may be removed with a pipette. They grow as buds or offsets from all parts of the internal surface of the vesicle. Many soon become detached from the wall of the cyst and die. Echinococci are represented in pl. LXXVI, figs. 2 to 10.

The echinococcus is developed from some of the masses of bioplasm, of which the inner wall of the cyst seems to be almost entirely composed. They may be seen at different stages of development in many cysts projecting like buds from the surface, pl. LXXVI, fig. 3. Two species of echinococci have been described, *E. hominis* and *E. veterinorum*, but it is probable that these are really the same.

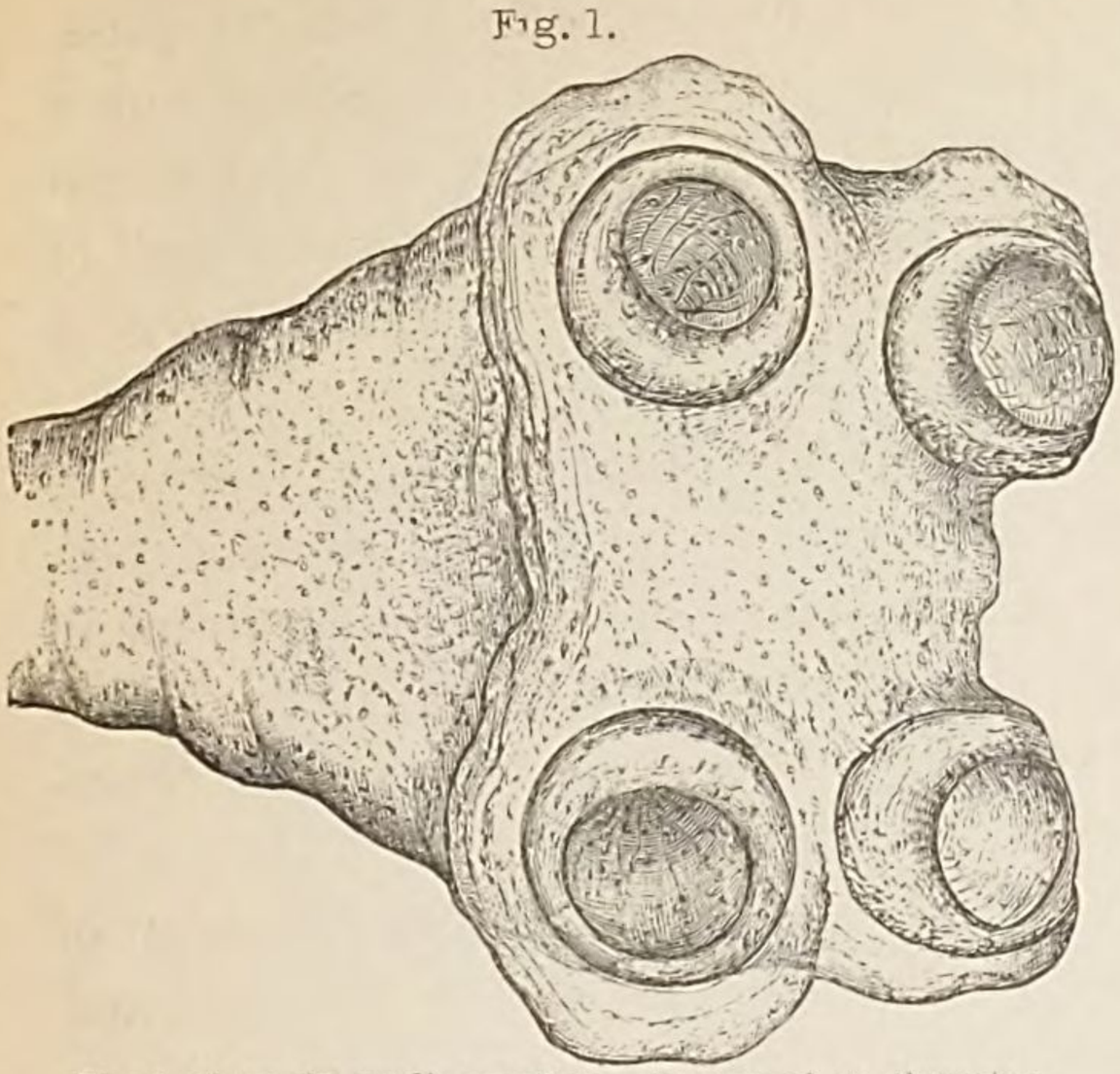
The echinococcus has been proved to be the immature condition of the *tænia echinococcus*, a minute tape worm found only in the dog and wolf. The tænia echinococcus is about $\frac{1}{4}$th of an inch long, and consists of only four joints, including the head, which has four suckers and a circle of hooks. The eggs contain a six-hooked embryo which does not develop into a cysticercus, but into a spherical vesicle containing a granular material. The echinococci are afterwards developed by budding from the inner wall of the vesicle. My friend and former pupil, Mr. Nettleship, has published some very interesting observations upon the development of this parasite in the Proceedings of the Royal Society for June 21st, 1866, and I have to thank him for some beautiful specimens of the tænia in the dog's intestine which were developed from the echinococci taken from the liver of a sheep. Some of the brood are represented in pl. LXXVI, fig. 3, and the tænia in fig. 2.

Fig. 4 represents the appearance of echinococci magnified with an inch object-glass, and in figs. 8, 9, are shown two specimens magnified with a quarter. In one of these the hooks are seen to be extruded, a condition which has been considered to result from the occurrence of endosmosis and commencing decomposition. They may be made to protrude their hooks by leaving the opened cyst for twenty-four hours in the fluid. The echinococcus is about 1-200th of an inch long. It is nourished by imbibition only. The hooklets are thirty-four in number.

If a little of the fluid from the interior be evaporated upon a glass slide, numerous crystals of chloride of sodium will be formed. Heintz gives the following plan for detecting *succinic acid* in the fluid of the hydatid cysts. The fluid concentrated by evaporation is to be treated with a little hydrochloric acid and agitated with water, and ether free from alcohol, until nothing more is taken up. The impure succinic acid obtained by evaporating the etherial solutions, is dissolved in water. The solution is to be filtered and evaporated to dryness. The residue is then treated with alcohol and completely recrystallized.*

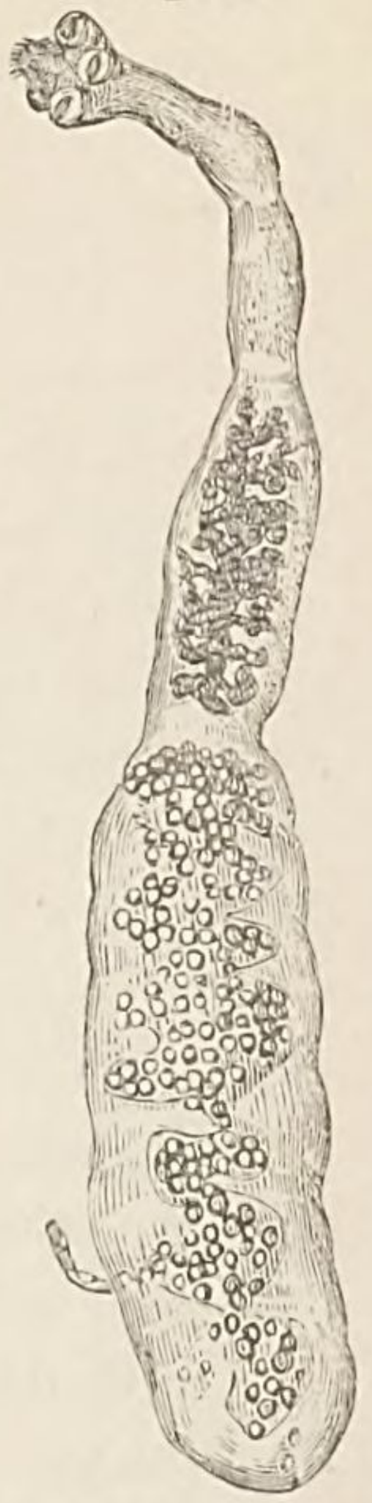
* Heintz, "Lehrbuch der Zoochemie," page 239.

Fig. 1.



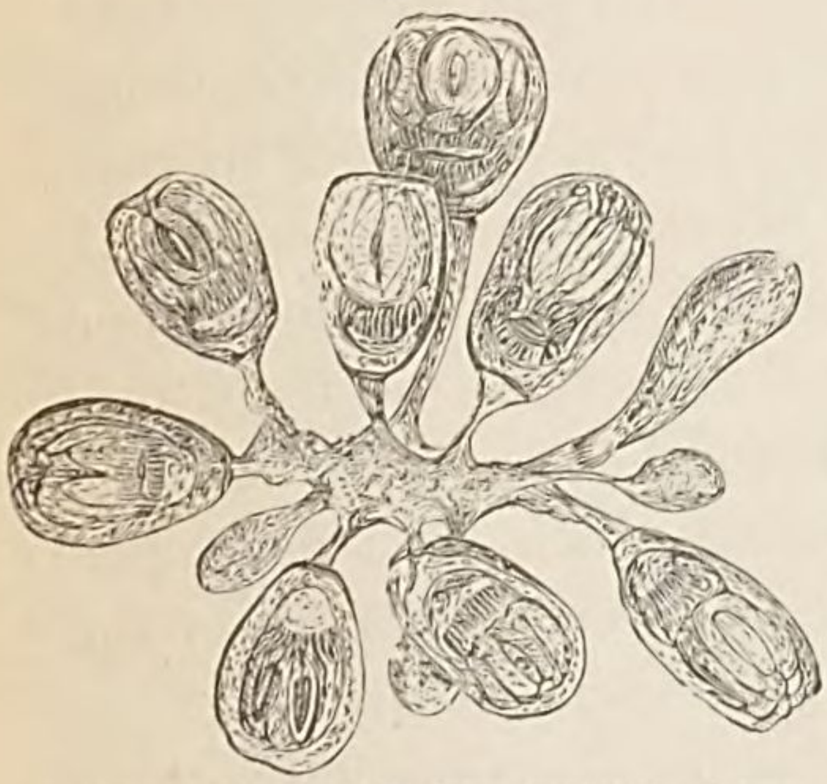
Head of tænia mediocanellata. Preserved in glycerine. × 20. p. 476.

Fig. 2.



Tænia echinococcus with ova. × 15. p. 478.

Fig. 3.

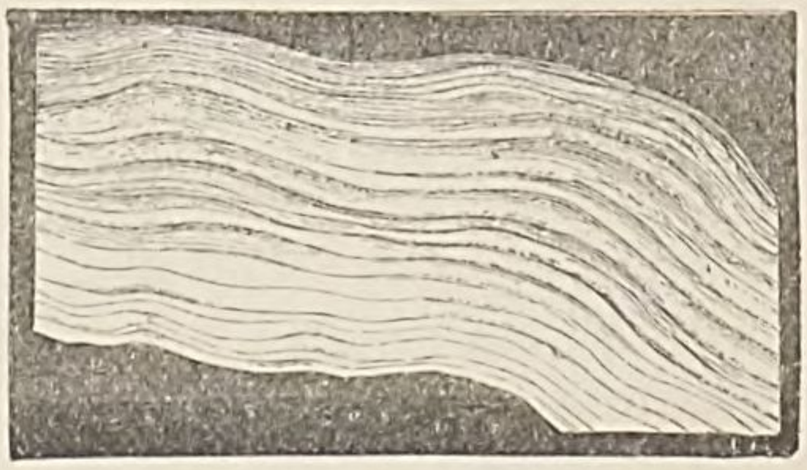


Group of echinococci from hydatid. Liver of ox. × 40. p. 478.

Fig. 4.

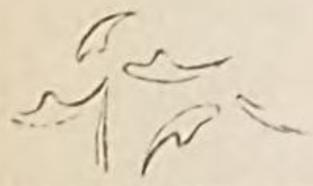
Echinococci. × 42. p. 475.

Fig. 7.



Layers of which the wall of an hydatid cyst is composed. × 215. p 477.

Fig. 5.



Free hooklets from echinococcus. × 215. p 479.

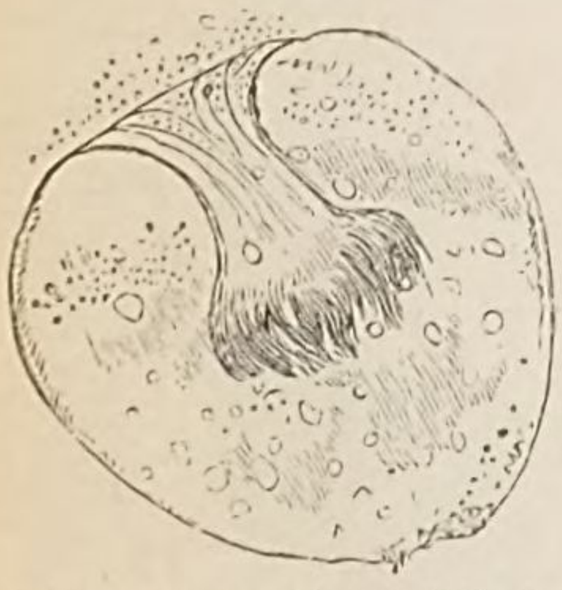
Fig. 6.

Hooklet of echinococcus. × 700. p. 479.

Fig. 10.

Hooklets of echinococci, masses of bioplasm, oil globules, crystals of fatty acid, and debris. Deposit from fluid of an hydatid cyst. × 215. p 479.

Fig. 8.



Echinococcus. Showing the head and suckers withdrawn. × 215.

Fig. 9.

Echinococcus with the hooks extended. × 215. p. 478.

1/100th of an inch |___| × 15.
1/1000th " _ × 40.

1/1000th of an inch |___| × 215.
" " |_____| × 700.

[To face page 478.

The character of the claws, pl. LXXVI, figs. 5, 6, 10, should be particularly noticed, as their presence is characteristic of echinococci. Hydatids are occasionally expectorated; usually in consequence of a cyst in the liver opening into the base of one lung. The appearance of the cysts in the sputum will direct attention to the origin of the pulmonary mischief, but the observation should be always confirmed, if possible, by the microscopical examination of the claws or hooks. Echinococci may be preserved in the creosote solution or in preservative gelatin. The hooks may be preserved moist in fluid, or dry in Canada balsam.

446. Trichina Spiralis is a species of entozoon which is sometimes found in the voluntary muscles. It was first described by Owen, in 1835. The researches of Leuckart and Virchow have proved that the Trichina spiralis is not the brood of the Tricocephalus dispar engaged in migration, as was formerly supposed. When introduced into the human intestine, the larval trichinæ rapidly arrive at sexual maturity, and ova are soon found and fertilised. The young filariæ soon after their escape from the uterus of the mother commence their migrations by boring in every direction through the surrounding tissues, till they arrive at the voluntary muscles. They rarely stop anywhere else. In the muscles they undergo development, and soon become surrounded by a cyst which eventually calcifies. It appears, however, that these cysts are by no means essential, and that they may be wanting even where the Trichinæ are very numerous, so that a careful microscopical examination would always be necessary to determine the absence of the parasite. Encysted trichinæ are represented in pl. LXXVII, figs. 1 and 2, in a portion of human muscle from a patient who died in the London Hospital. I am indebted to Mr. Curling for a specimen of the muscle. A figure of the trichina removed from the cyst is given in the same plate, fig. 4. Fig. 3 is a very good drawing given me by my friend Professor Brown, of the veterinary department of the Privy Council. This entozoon has acquired a peculiar interest of late in consequence of the fatal results which have occurred in many cases in Germany, from eating flesh infected with trichinæ. The trichina disease usually commences with diarrhœa or abdominal pains, symptoms which are caused by the presence of the larvæ in the intestines. The next symptoms are those which result from the invasion of the muscles by the parasite, and consist chiefly of great prostration, severe pain in the muscles, and inability to extend the limbs, forcible extension being accompanied by great pain. The patient's face and limbs become œdematous, and there is much fever. Recovery from this stage is sometimes followed by debility and wasting, and death may result from the impaired capacity for exertion of the muscles consequent on the presence of the Trichina cysts in them. The disease is not in-

variably fatal. It is remarkable that the very same species of Trichina will flourish in a number of very different classes of animals, and is found in birds as well as in mammals.

The calcification of the cysts renders them opaque, so that the contained worm cannot be distinctly seen till the calcareous matter has been dissolved out by dilute hydrochloric acid, pl. LXXVII, fig. 2. The worms may die in the capsules which have undergone calcification, and not only may the hard calcareous matter be absorbed, but even the fat vesicles which are developed in considerable number at each end of the capsule may eventually disappear. A very full account of the Trichina will be found in Leuckart's work, vol. ii, p. 509. See also the ("Medico-Chirurgical Transactions," vol. xlvii, p. 55), memoirs by Dr. Althaus and Dr. Thudichum.

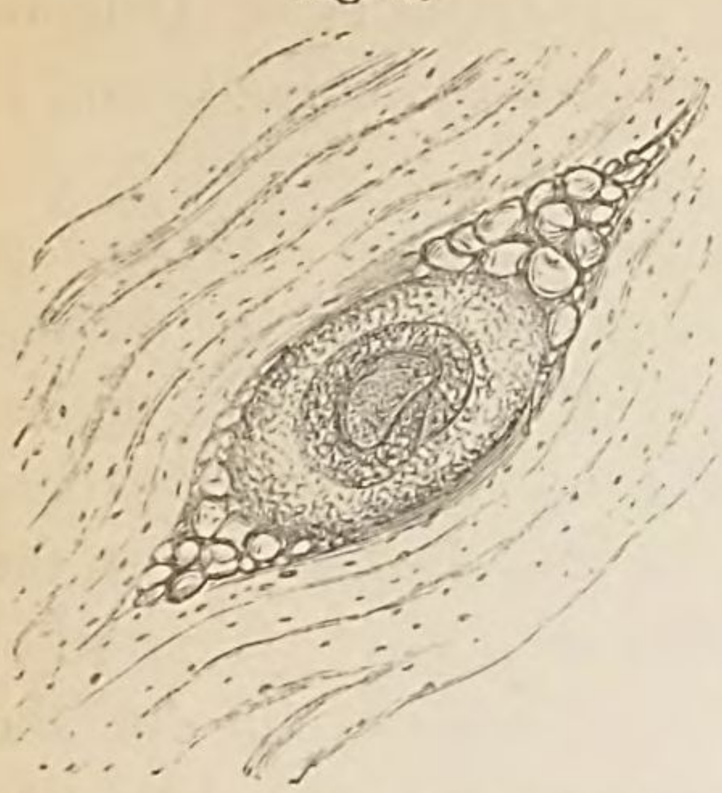
441. Filaria or Hæmatozoon of Human Blood (Lewis).—Two of the most interesting and important discoveries recently made by the use of the microscope are:—1. The discovery of a *Filaria* in chylous urine, and 2, the discovery of the same worm in human blood. In March, 1870, Dr. Lewis, Her Majesty's British forces, Calcutta, found in a specimen of chylous urine some very minute worms. Upon examining other examples of chylous urine, more filariæ were found, and Dr. Lewis states that not in a single case that came under his notice were the worms absent. Through the kindness of my friend Dr. Palmer, I received from Calcutta some of these worms in a tube. They arrived in good preservation, and the drawings, fig. 5, pl. LXXVII, and fig. 1, pl. LXXVIII, were taken from them. So far from the worms being injured by their long journey, it will be observed that one or two points of structure not clearly indicated in Dr. Lewis's drawings, were distinctly evident in these specimens.

In 1872, Dr. Lewis was fortunate enough to make the further discovery of multitudes of these same filariæ in the blood of several patients suffering from chylous urine. There can be no longer any doubt concerning the nature of this hitherto obscure affection. The filariæ after causing obstruction in the capillaries make their way through the stretched distended vascular walls into lacteal and lymphatics, and growing and multiplying in these channels form obstructions, in consequence of which extravasations occur and in many instances communications are opened between some part of the lacteal system and the urinary organs.* Chyle, and with it many of the filariæ, find their way into the urine. Probably many obscure symptoms occurring in this and allied affections will be explained ere long if these interesting discoveries are carefully followed up.

442. Method of detecting the Filariæ in the Blood.—In order to

* "Kidney Diseases, Urinary Deposits, and Calculous Disorders," 3rd edition, page 307.

Fig. 1.

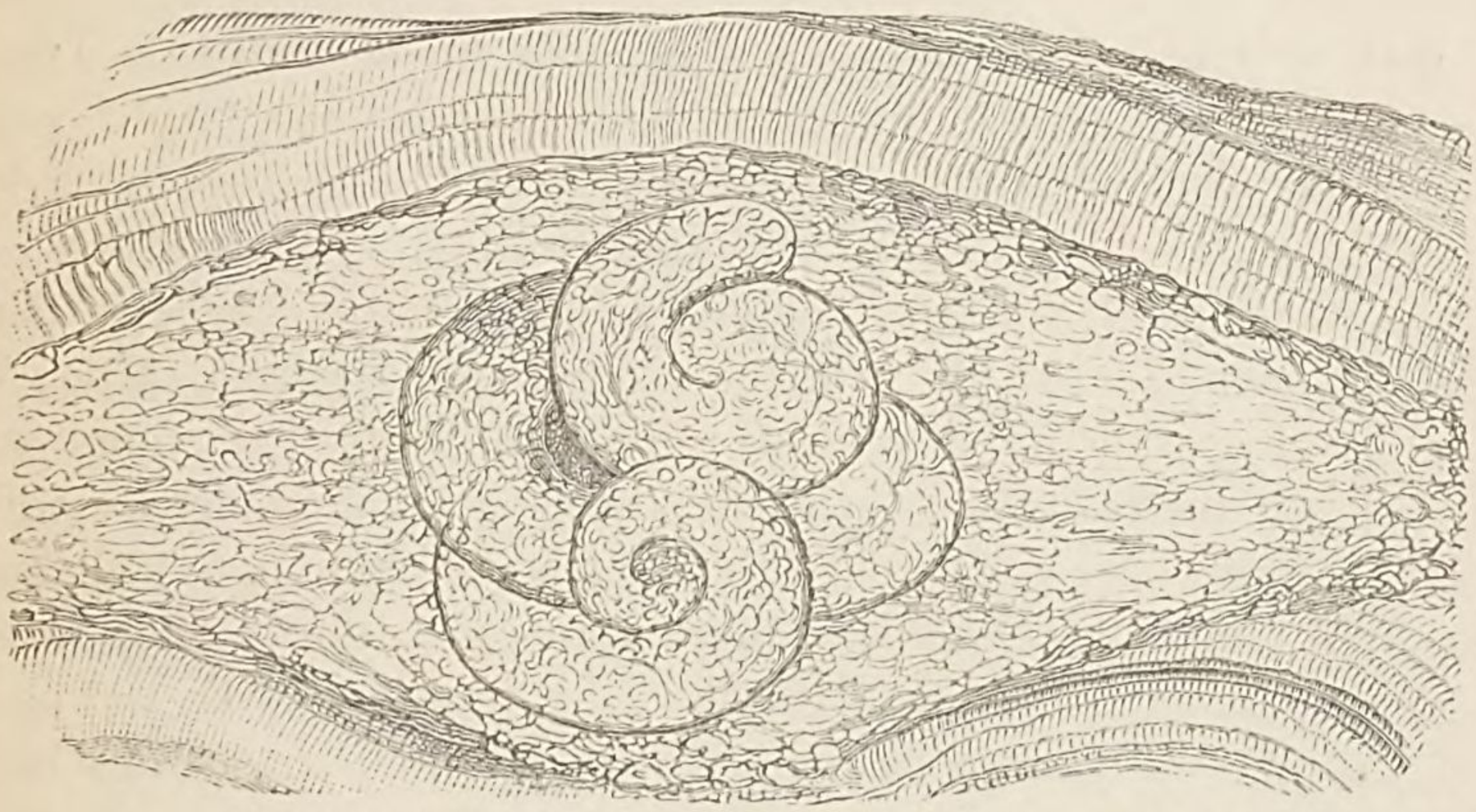


Trichina spiralis, with capsule subject. Mounted in glycerine From human × 40 p. 479.

Fig. 2.

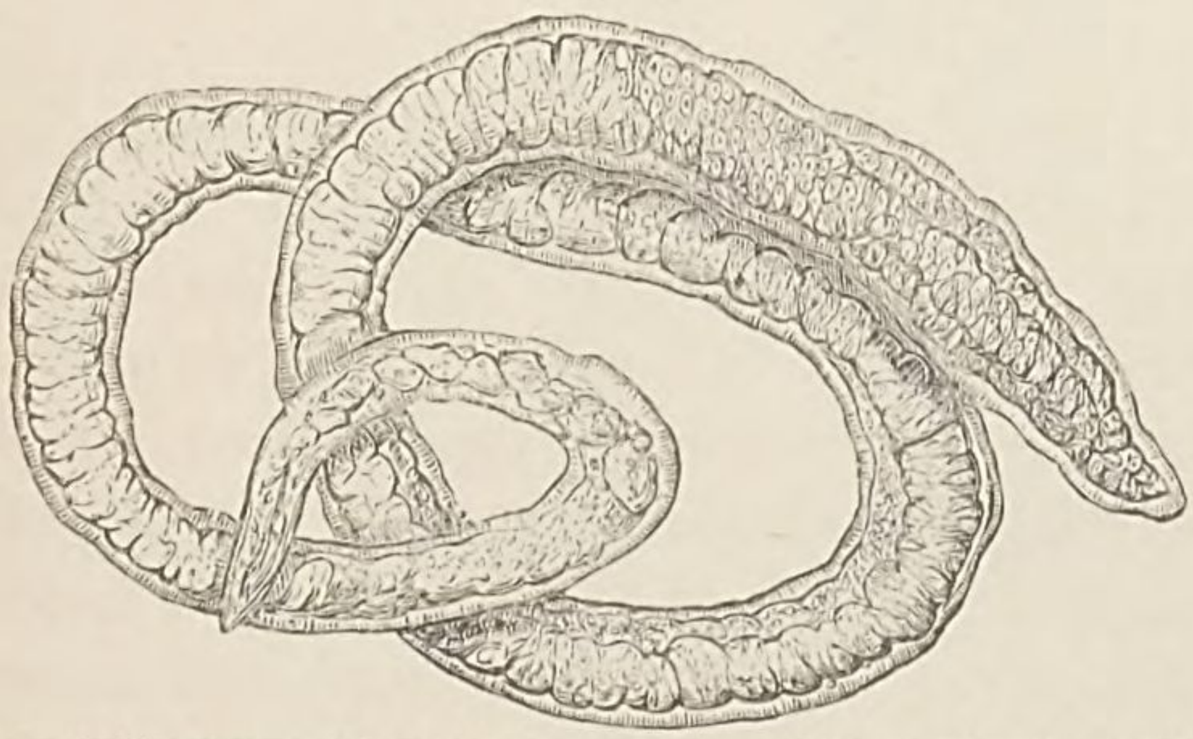
Capsule containing two trichinæ. From human subject. × 130. p. 480.

Fig. 3.



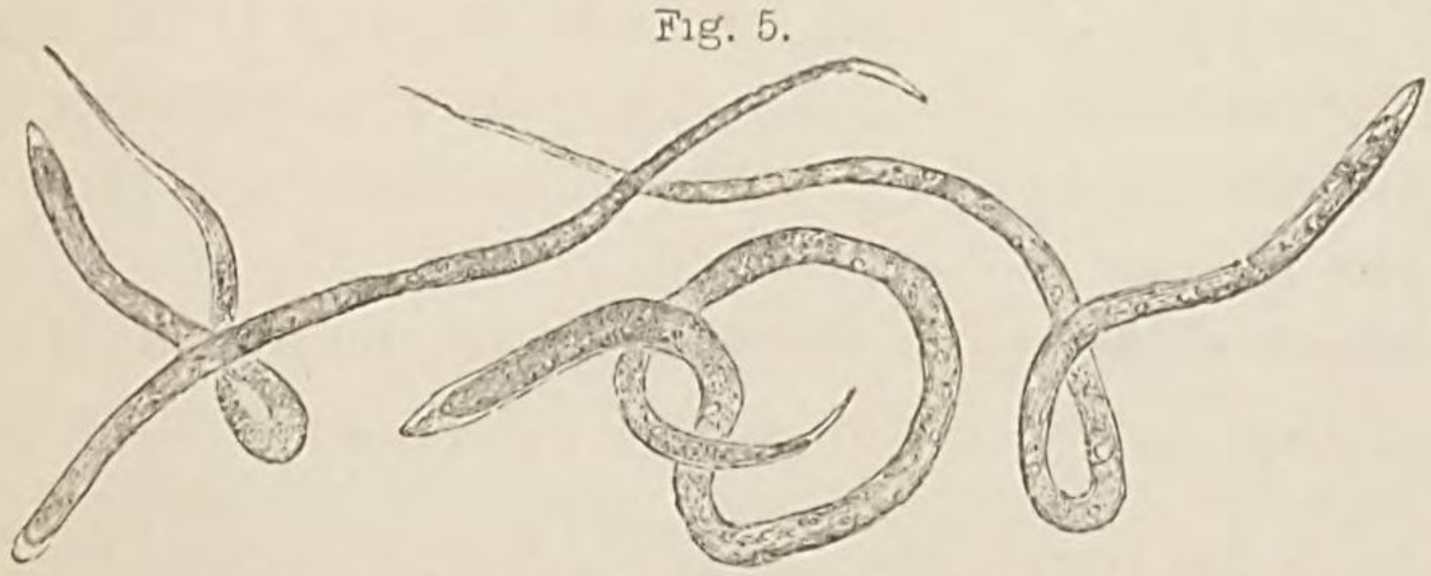
Trichina spiralis in muscle. From a drawing by Prof. BROWN. p. 479.

Fig. 4.



Trichina spiralis, removed from its capsule. From human subject × 215. p. 479.

Fig. 5.



Filaria or Hæmatozoon, from a specimen of chylous urine. The worms were collected by Dr. PALMER, of Calcutta, and sent to me by post in a small glass tube. × 215. p. 480.

1000th of an inch ⎵ × 40.
" " ⎵⎵ × 130.
" " ⎵⎵⎵ × 215.

[To face page 480

detect the Hæmatozoon during life, Dr. Lewis adopts the following method :—"A piece of narrow tape is coiled tightly round the end of one of the fingers or toes, so as to produce a little temporary local congestion of the part, but not to such an extent as to cause the slightest pain; and with a clean, finely-pointed needle the finger is gently pricked—half-a-dozen slides and covering glasses having been previously prepared. The drop of blood thus obtained will suffice for several slides, but I find it a good plan to squeeze out only a very small drop, and transfer it altogether to one slide by drawing the edge of the covering glass along the tip of the finger so as to *scrape* off the 'droplet.' The glass is then carefully pressed on to the slide by a gliding motion, in order to produce as thin a layer of fluid as possible, and to ensure that all the fluid removed is retained beneath the cover, because there is a tendency on the part of the fluid to carry the Hæmatozoa towards the edge of the slide, just as is observed to take place in examinations of the urine for 'casts' of the kidney-tubules.

"The slides are now to be carefully and systematically examined; a lateral and horizontal stage-movement being very useful for this purpose, as it enables us to make sure that every part of the preparation has been scrutinized.

"It must not be supposed that the filariæ are to be detected by taking a mere peep through the microscope; sometimes, certainly, I have observed one, or two even, in the first field examined; but this is by no means usual, and their detection often demands considerable patience. Each slide will require about a quarter of an hour before it can be satisfactorily explored; any one who imagines that they can be detected with the same ease as a white-blood corpuscle had better not make the attempt.

"Several slides may have to be examined, and it may be necessary to make a fresh puncture, for I have found the Hæmatozoa to be absent in several slides obtained from one finger, but present in all the slides obtained from another at the same time; whereas on making a fresh puncture in the finger where none had been found at first, it was ascertained that they were equally numerous in both. This is possibly accounted for by the little orifice made having become plugged by fat, &c., so that the blood squeezed through had to some extent been filtered, for although this microscopic filaria can pass through any channel permeable to a red-blood corpuscle, still, when it is considered that the length of the former is nearly fifty times its diameter, the wonder is that they are not more completely prevented from escaping through so fine an orifice even when perfectly patent.

"The search should not be undertaken with too high a magnifying power, but it should be sufficiently high to define the outline of a red-blood corpuscle quite distinctly. I have found that a good two-thirds

of an inch objective answers the purpose of a *searcher* admirably; it embraces a tolerably large area, so that the preparation can be examined in a comparatively short time; but care should be taken to keep the fine-adjustment screw constantly moving, so as to examine the deeper as well as the superficial layer of fluid in each field as it passes under observation. Should anything unusual be observed, the low power must be replaced by a $\frac{1}{4}''$ or, better, a $\frac{1}{8}''$ objective.

"In order to keep the active hæmatozoon under observation for some hours, a camel-hair pencil dipped in a solution of Canada-balsam or mastic in chloroform, should be passed along the edges of the covering glass, so as to prevent evaporation, and the formation of air spaces in the preparation."*

442. Movements of the Filaria.—Dr. Lewis describes the movements of the living entozoon as follows:—"At first the movements of the hæmatozoon were so rapid that little could be detected in addition to what had been quite as distinctly seen with $\frac{1}{4}''$ glass, except that in certain positions assumed by the worm, and in certain lights, extremely fine transverse striæ were observed quite distinctly. The existence of these striæ had, on several occasions, been more than suspected under the lower power ($\frac{1}{4}''$), but they could not be satisfactorily demonstrated. No attempt has been made to represent those fine markings in the woodcuts as seen by such a, comparatively, low power as this, for it would only tend to mislead; to cut lines in wood only $\frac{1}{25,000}$ of an inch apart (which is about the distance between the markings), when simply magnified 300 diameters, would be impossible, and even in the engraving which represents the object as magnified by twice this power, the distinctness of these markings is considerably exaggerated." See drawing, fig. 1, pl. LXXVIII, p. 484.

"As the movements of the filaria become a little slower, it was seen that the striæ were not on its outer coat, but confined to the body of the worm, and that the tail, which almost always under the $\frac{1}{4}''$ objective looked like a lash, was not so in reality, but that, every now and then, it could be seen flapping against the corpuscles like a fin—sometimes vertically, sometimes horizontally, and then becoming folded upon itself like a ribbon, a condition which I had already observed and figured two and a half years ago without knowing what it was. Precisely similar phenomena were observed to occur at the opposite terminal extremity.

"It was, however, only after the lapse of fully five hours' careful watching, the activity of the hæmatozoon having considerably subsided, that the real nature of what appeared to be the rapid protrusion and retraction of the delicate membrane at the oral and caudal terminations

* "On a Hematozoon inhabiting Human Blood; its relation to Chyluria and other Diseases." Calcutta, 1872. Forming an Appendix to the eighth Annual Report of Sanitary Commissioner with the Government of India.

was discovered. An unusually long tail was seen to be trailing after the 'body' of the filaria for several seconds, and whilst thus being dragged, fortunately, it remained exactly in focus, when suddenly the ribbon-like folds were straightened by the darting of the pointed extremity of the worm into the very tip of this hyaline filament. Scarcely had this taken place than the tail was again retracted and the ribbon-like appendage became evident once more; whereupon the ribbon-like filament at the other extremity was suddenly straightened in a similar manner, and the 'head' rapidly projected into the very tip.

"The hæmatozoon may, therefore, be said to be *enveloped in an extremely delicate tube, closed at both ends*, within which it is capable of elongating or shortening itself. This tube, like the sarcolemma of muscular fibres, is without any visible structure, is perfectly transparent, and, but for the difference between it and the fluid in which it is immersed in its power of refracting light, which allows of its margins or folds being brought into view, it could not be demonstrated.

"The fact of its being thus enclosed seems to show that in the present stage of its existence, the 'home' of this filaria is in the blood; it has no visible means of perforating the tissues; moreover, although constantly observed to be in a state of great activity, it does not seem to manifest any special tendency to migration, and is apparently dependent on the current of the blood for its transference from place to place; its movements, therefore, within this enveloping tube, appear to be as limited as those of any other animal enclosed within a sac."*

443. Adult form of the Filaria of Human Blood.—In the spring of 1876, Dr. Bancroft of Brisbane, confirmed the observations of Dr. Lewis, and forwarded some blood containing filariæ to England. These were examined by Dr. Cobbold, who begged Dr. Bancroft to continue his researches, and suggested that the adult form of the worm should be "sought for in the human bearer." The following is Dr. Bancroft's communication to Dr. Cobbold, dated Brisbane, Queensland, April 20th, 1877, and published in the "Lancet" of July 14th, 1877:—

"I have laboured very hard to find the parental form of the parasite, and am glad to tell you that I have now obtained five specimens of the worm, which are waiting to be forwarded by a trustworthy messenger.†

"I have on record about twenty cases of this parasitic disease, and believe it to be the solution of chyluria, some forms of hæmaturia, one form of spontaneous lymphatic abscess, a peculiar soft varix of the groin, a hydrocele containing fibrinous fluid, another containing chylous fluid, together with some forms of varicocele and orchitis. These I have verified.

"In the colony there are no cases that I can find of elephantine leg,

* Op. cit., page 25.

† The adult worm was described and figured by Dr. Lewis in 1874.

scrotal elephantiasis, or lymph scrotum; but, from the description of these diseases in the volume on Skin and other Diseases of India by Fox, Farquhar, and Carter, and from Dr. Roberts's article on the latter in his volume on Urinary Diseases, I am of opinion that the parasitic nature of the same will be established.

"The worm is about the thickness of a human hair, and is from three to four inches long. By two loops from the centre of its body it emits the filariæ described by Carter in immense numbers.

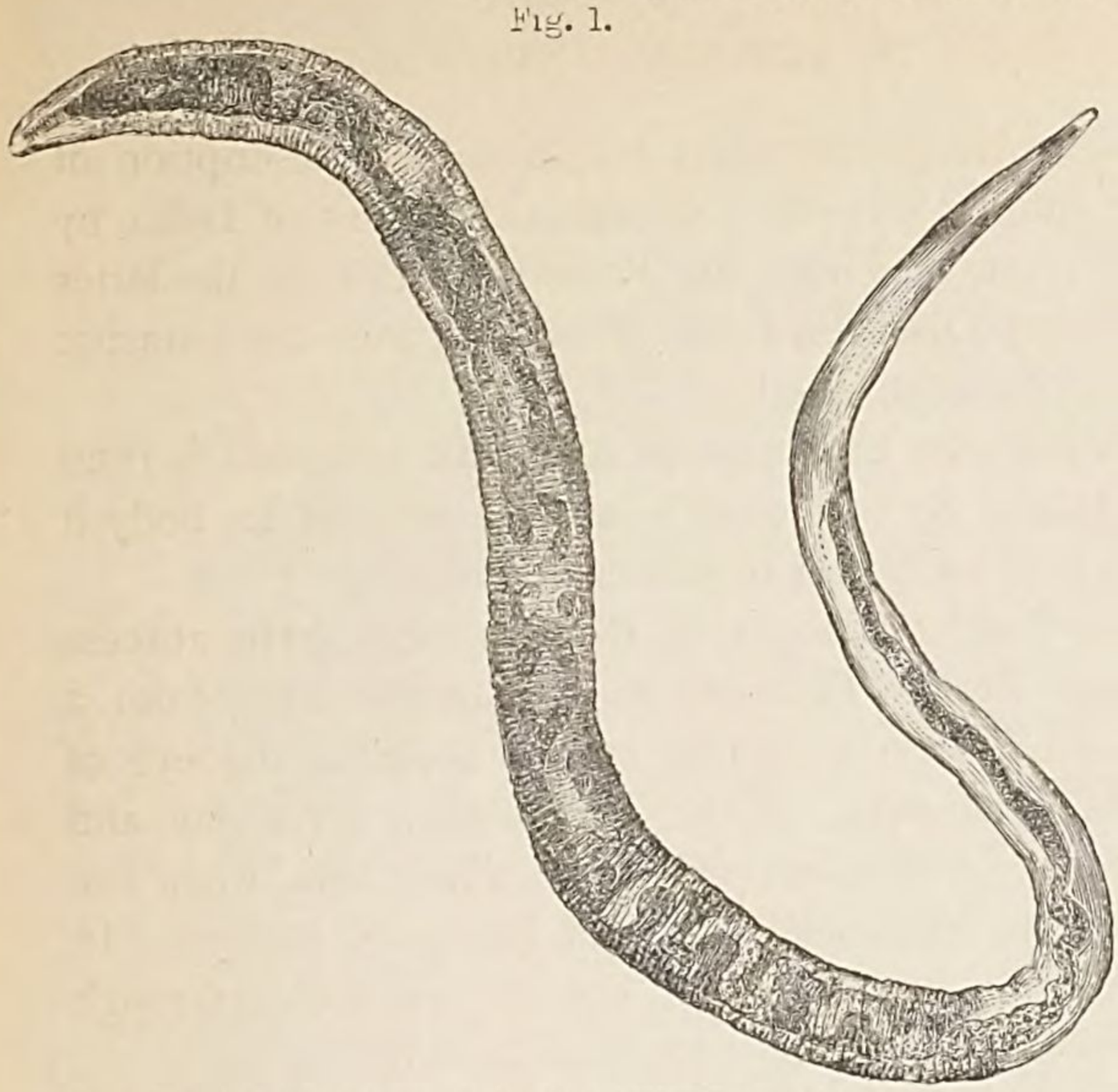
"My first specimen I got on Dec. 21st, 1876, in a lymphatic abscess of the arm. This was dead. Four others I obtained alive from a hydrocele of the spermatic cord, having caught them in the eye of a peculiar trocar I use for tapping. These I kept alive for a day, and separated from each other with great difficulty. The worm, when immersed in pure water, stretches itself out and lies quite passive. In this condition it could be easily washed out of hydroceles through a large-sized trocar from patients known to suffer from filariæ.

"I will forward you full particulars of my cases (and the worms) at an early date." This seems to complete a most important series of observations. Dr. Cobbold proposes that this newly discovered nematode of the human subject, the probable adult form of the minute filariæ discovered in chylous urine and in blood by Dr. Lewis, should be called *Filaria Bancrofti*. For further observations on this filaria, *see* page 503.

444. Distomum or Bilharzia Hæmatobium.—Dr. John Harley recently, 1864, called attention to the existence of endemic hæmaturia in certain parts of the Cape of Good Hope, and in Natal, and showed it to be due to a species of Bilharzia which, after careful comparison with Griesinger's figures of Bilharzia hæmatobium, he has been induced to refer to another species named by him, B. Capensis ("Medico-Chirurgical Transactions," vol. xlvii, page 55). As no opportunity has yet been afforded of examining the adult animal, it is, however, probable that the species is identical with B. Hæmatobium.

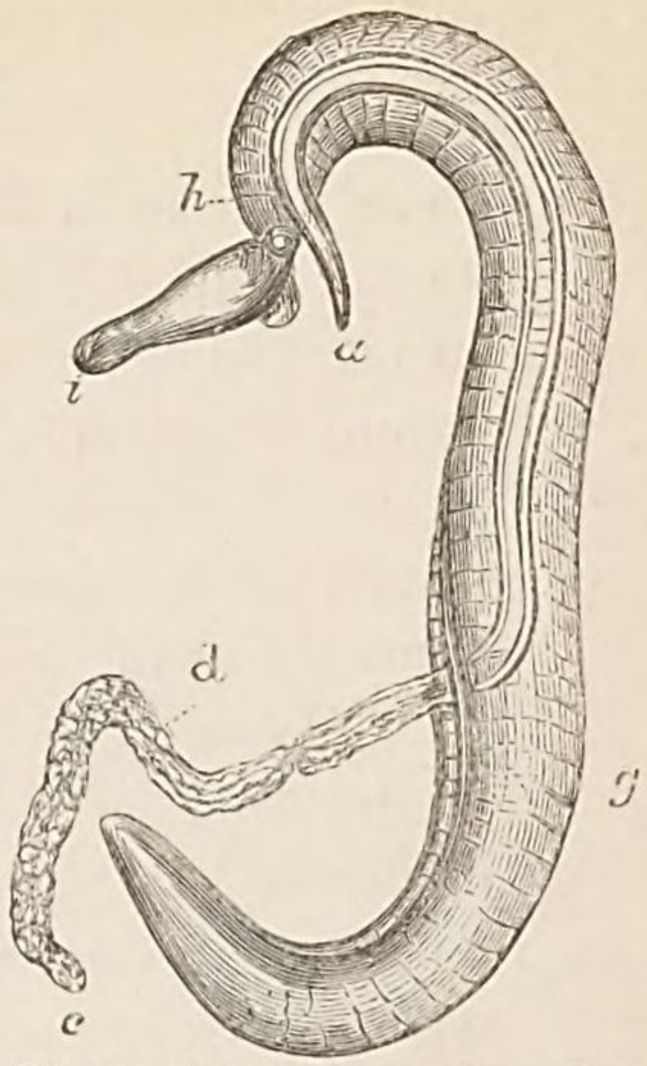
Dr. Harley found in the urine of his three patients the eggs and ciliated embryos of the parasite, also part of its intestine, and a portion of ciliated integuments. Drawings of the eggs embedded in mucus and free, are given in pl. LXXVIII, figs. 13, 14, and 15. For the specimens from which these drawings were made, I have to thank my friend and former colleague. This parasite is a non-hermaphrodite trematode worm. It has two suckers, and in the body of the male is a peculiar channel, the "gynæcophoric canal," which contains the female during copulation, fig. 12, pl. LXXVIII. The parasite is found in the vesical, mesenteric, and portal veins, and by its presence in their minute branches gives rise to lesions of the mucous membrane of the intestines, bladder, ureters, and pelves of the kidneys. The principal symptoms

Fig. 1.



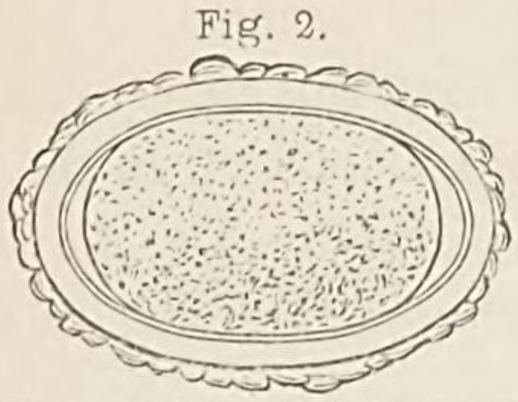
One of the worms represented in plate LXXVII, magnified 700 diameters. p. 480

Fig. 12.



Bilharzia hæmatobium. (After BILHARZ) *h, i, g,* male, with gynecophoric canal in which the female *a, b, c,* lies during copulation. p. 484.

Fig. 2.



Ovum of ascaris lumbricoides. After LEUCKART. × 370. p. 485.

Fig. 13.

Ova of bilharziahæmatobium in urinary deposit. Drawn from a preparation of Dr. HARLEY's. × 15 p. 484.

Fig. 3.

Distomum lanceolatum. × 370. p. 485.

Fig. 4.

Tænia mediocanellata. After LEUCKART × 37. p. 476

Fig. 5.

Tænia solium. × 370. p. 476.

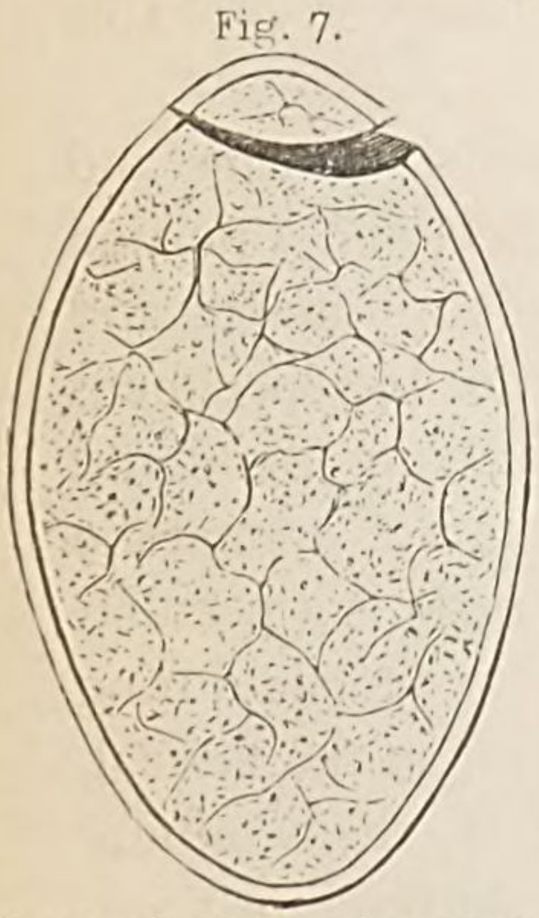
Fig. 6.

Bothriocephalus latus. p. 476

Fig. 14.

Ova of bilharzia hæmatobium from urine. Drawn from Dr. HARLEY's preparations. × 130. p. 484.

Fig. 7.



Ovum of distoma hepaticum. After LEUCKART. × 370. p. 485.

Fig. 8.

Ovum of bothriocephalus latus × 370.

Fig. 9.

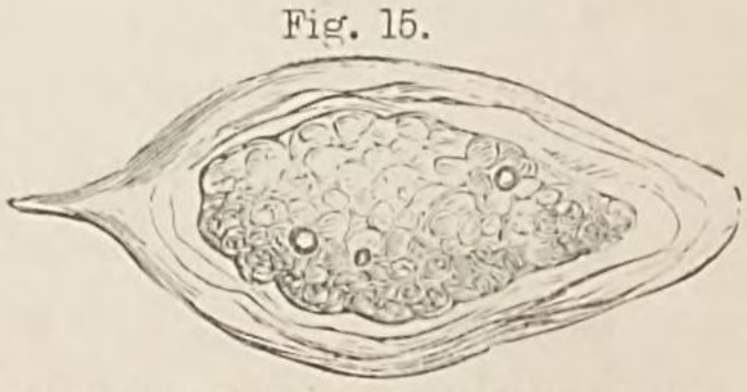
Oxyuris vermicularis. × 215. p. 485.

Fig. 10.

Fig. 11.

Ova of trichocephalus dispar. After LEUCKART. × 370. p. 485.

Fig. 15.



Ovum of bilharzia hæmatobium. × 215. p. 484.

1000th of an inch ⌣ × 40.
, ,, ⌣ × 130.
,, ,, | | × 215.

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are diarrhœa and hæmaturia, accompanied by great anæmia and prostration of strength. After a certain time the ova and embryos of the parasite are found in the urine. Dr. John Harley considers that the eggs often become, after the total disappearance of the hæmaturia, the nuclei of renal calculi. After death the mucous membranes affected are found studded with extravasations of blood, and more or less thickened and ulcerated.

445. Other Entozoa.—The common *fluke* (*Fasciola* or *Distomum hepaticum*) forms a very interesting object for examination. One species may generally be met with in the bile-ducts of the ox and sheep. The highly branching digestive and water vascular systems may be injected with different colours—the former through the oral opening in the anterior sucker, the latter through the caudal aperture of the aquiferous system. The male and female organs of generation should also be studied.

The small thread worms *oxyuris vermicularis* are common in children, and are met with chiefly in the rectum. The *tricocephalus dispar* is met with in the cœcum and colon. The *ascaris lumbricoides* or great round worm, is usually found in the small intestine.

Ova of several of the most common entozoa are represented in pl. LXXVIII, figs. 2 to 11. An explanation is appended to each figure.

The only other entozoon which need be alluded to here is the *Strongylus gigas*, the largest of the entozoa. This is very rarely met with in man, but is not unfrequently seen amongst animals. It is usually found in the kidney. Some years ago I met with three of these creatures, two males and a female, coiled up in what had been the kidney of a dog, but which was reduced to a thin membranous cyst. The ureter was quite pervious, and the mucus on the surface of its mucous membrane, with that of the bladder, contained very numerous ova. For microscopical examination of the tissues of this creature, it must be dissected under water. The intestine is square, and contains altered blood. The ova form beautiful objects.

446. Entozoon-like Bodies in the Muscles.—In the muscular fibres of the heart, and less frequently in the voluntary muscles of many of the animals killed for food, are found some peculiar living bodies which have long been known to observers, although their nature has not yet been determined. They were discovered in 1843 by Miescher in the muscles of a mouse. Hessling found them in the muscular fibres of the heart of the sheep and ox. Siebold and Bischoff demonstrated them in the rat and mouse, and they have been also found in the deer. In 1855, Rainey found and figured similar bodies from the muscles of the pig, and by him they were supposed to be the cysticercus cellulosæ in an imperfectly developed state. "On the structure and development of the Cysticercus Cellulosæ, as found in the muscles of the pig," Phil.

Trans., vol. 147, p. iii, 1857. They have since been often termed Rainey's bodies. The observations of later observers have not confirmed the conclusions of the last-named writer. Although it is not known what these unquestionably parasitic bodies really are, it is quite certain that they have nothing to do with the cysticercus cellulosæ. These bodies cannot be properly included with intestinal worms, but upon the whole this is, perhaps, the most convenient place for considering their structure.

In my investigations upon the muscles of animals destroyed by the cattle plague, I found these bodies in enormous numbers. While they are ordinarily found largely in the muscular fibres of the sheep's heart, and to a less extent in that of the ox; they are not to be detected in the best beef and mutton. On the other hand in almost every specimen of cattle plague beef which I examined, these entozoon-like bodies were present, and in many cases, in immense numbers. Moreover, the bodies found in the systemic muscles attain a size and degree of development seldom, if ever, observed in those found in the heart.

As further observations upon this subject are much required, I feel that it is desirable to call attention to it in this place. In plates LXXIX and LXXX, I have repeated several of my figures from my report on the Cattle Plague, 1866. The conclusions I arrived at may be summed up as follows :—

1. That in almost all, if not in all, animals dying of Cattle Plague, entozoon-like bodies exist in considerable number in the voluntary muscles of the system and in the heart.

2. They are occasionally found, but in comparatively small numbers in animals apparently in perfect health when killed.

3. These or closely allied species have been known for more than twenty years, but their nature has not yet been determined. They have been found in the ox, sheep, deer, pig, rat, mouse, and perhaps other animals.

4. In the muscles of a calf killed by Cattle Plague, under *six months* of age, these bodies were found in immense numbers.

5. They vary in length from less than the $\frac{1}{300}$th of an inch to at least a quarter of an inch in length. They are, for the most part, imbedded in the contractile material of the elementary muscular fibre, but they are occasionally found free.

6. They are for the most part spindle-shaped, and the external investment or envelope exhibits a very beautiful and peculiar structure being completely covered with delicate hair-like processes.

7. The mass within appears granular to low powers, and exhibits a division into numerous segments, but it is found to consist entirely of minute bodies resembling one another, possessing very definite characters, less than $\frac{1}{2000}$th of an inch in their longest diameter, and of

Fig. 1.

Fig. 2.

Fig. 3.

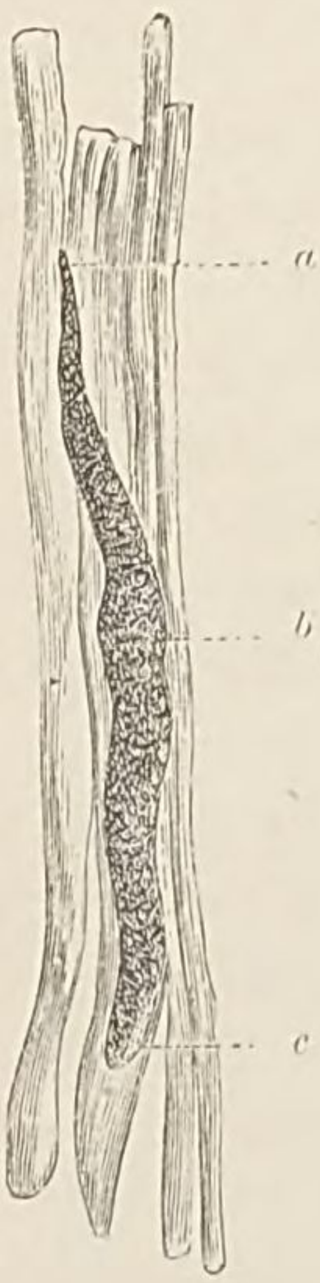
Fig. 4.

a

Worm like bodies of different sizes. From the voluntary muscles. Cattle plague. The largest, Fig. 1, *a*, is about the eighteenth; the smallest, Fig. 4, less than the one-hundredth of an inch in length. × 25. p. 486

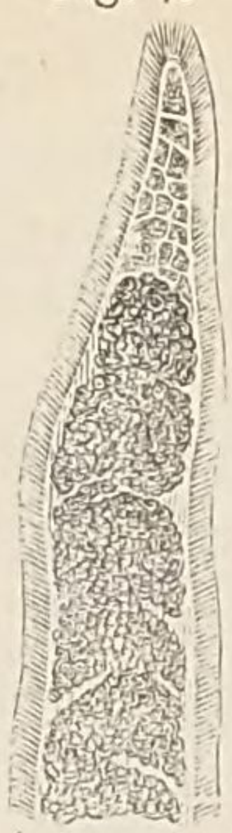
Fig. 5.

Fig. 6.



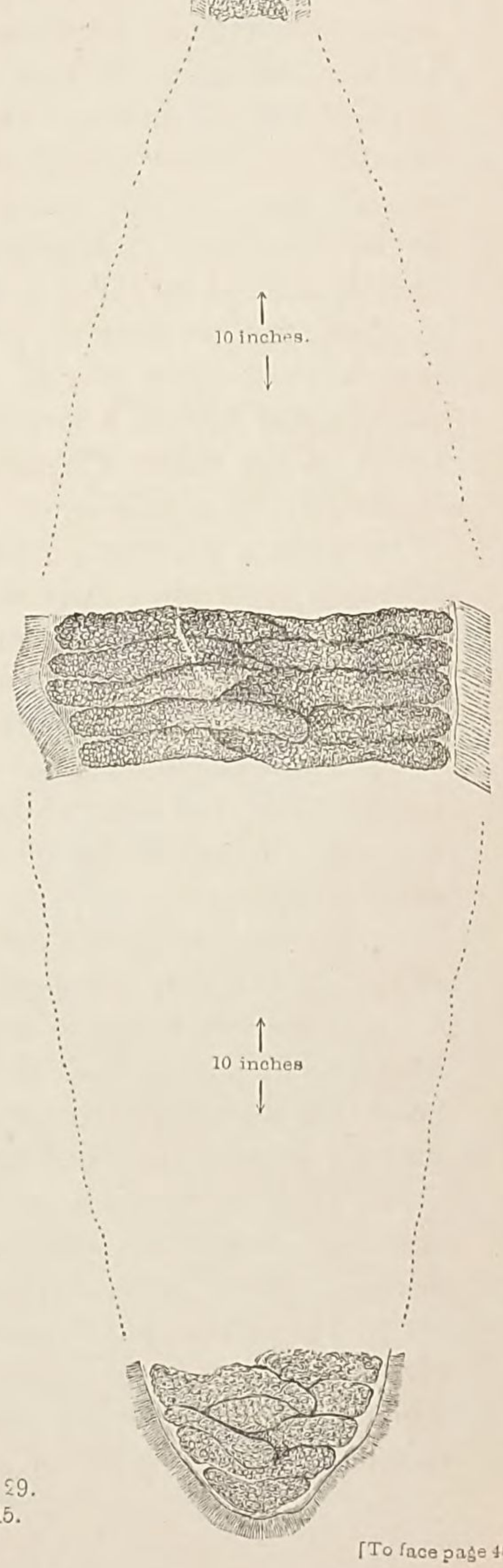
Worm like bodies from elementary muscular fibres, from the voluntary muscles. Cattle plague × 25. Fig. 6 is more highly magnified in Fig 7, the portions marked *a*, *b*, *c*, being alone represented. p. 486.

Fig. 7.



a

The upper, middle, and lowest portions of the same worm-like body as that represented in Fig. 6, but magnified 215 diameters. The division of the contents into collections, apparently the result of segmentation of the mass at an earlier period, was very distinct. p. 486.



100th of an inch |_____| × 29.
1000th of an inch |___| × 215.

L.S.B., 1866.

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peculiar form, being oval, flattened, the body slightly curved laterally, with one extremity blunt, and the other almost pointed, pl. LXXIX. This figure will enable the reader to form a notion of the great size of these bodies. Were this drawing completed it would be two feet in length; the real worm-like body being about the eighth of an inch long.

8. The entire mass increases in size as these small bodies increase in number, probably by division and subdivision within the cyst.

For further information upon this subject, and other drawings of the bodies in question, the reader is referred to the "Medical Times and Gazette" for 1866, the Cattle Plague Report, a paper published in the "Popular Science Review," No. 19, April, 1866, page 153, and Prof. Gamgee's work on the Cattle Plague; and for a more complete account of the literature of the subject, *see* a paper by Dr. Cobbold in the "Lancet" for January 27th, 1866.

Leuckart gives a most imperfect account of these bodies without any figures of them: indeed he almost contents himself with showing that they have nothing to do with the trichina, a conclusion with which everyone who has studied the two organisms with any care will cordially agree. It is extraordinary that these remarkable bodies have not received the attention from observers which they unquestionably deserve. My own observations in 1866 have been little noticed. The bodies in question are most remarkable. The figures in plates LXXIX, LXXX, are careful copies from specimens, and I hope some day they may be brought under the notice of Leuckart and other Helminthologists. Whatever may be the nature of the bodies in question, I can affirm that the representations I have given of them are accurate. The structure of the enveloping membrane would lead one to suppose that the worm-like organism was an early stage of the development of some entozoon. They may be related to the gregarinæ, but they differ remarkably from any psorospermiæ, among which they are generally included.

Peculiar entozoa are occasionally met with in the systemic muscles of the frog, newt, toad, and other animals, and it is very probable that by more careful and detailed investigation, the number of parasitic bodies inhabiting the voluntary muscles will be largely increased.

447. Examination of Entozoa.—The microscopical examination of entozoa does not usually present any great difficulty. The smaller species may be examined entire in the usual way, but the larger ones require dissection, and as the structures are often very delicate, the operation had better be performed under water, after the creature is quite dead and muscular contractility has passed off.

Many entozoa are preserved very satisfactorily in glycerine. I have some beautiful preparations of flukes mounted in this medium which

have retained their characters for several years. The process of staining with carmine and preserving in strong glycerine, described in Chapter VII, is well adapted for investigations on the entozoa. Entozoa may be mounted in preservative fluids, or dried and placed in balsam.

Protozoa.

Many of the protozoa may be included among the parasites. Of the so-called infusorial animalcules there are several species found in man himself which have been identified, and there are some minute organisms which probably pass part of their life in some of the lower animals or plants, and reach another phase of development in man.

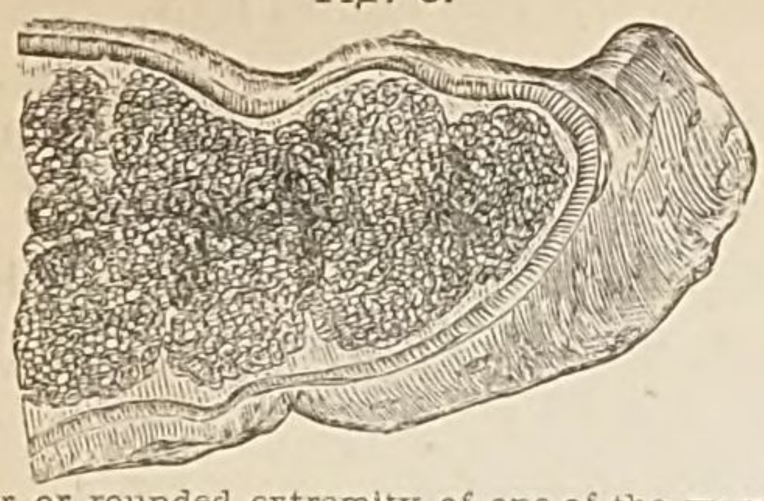
Of monads, Davaine distinguishes two varieties. *Cercomonas urinarius* and Trichomonas vaginalis of Donné and some others have been named. Various forms of *Paramecium* have been found in the cœcum and large intestine, and bodies which are probably allied to amœbæ have been met with in many situations. These must not be mistaken for particles of bioplasm belonging to the body itself, which, as is well known, exhibit movements (amœbiform).

Vegetable Parasitic Organisms.

There are many vegetable organisms of simple structure and of a low degree of organisation, which not unfrequently fall under the notice of the practitioner. Some are found growing upon the surface of the skin or mucous membrane in certain forms of disease, and in the recent fluid secretions. Others again are developed after the secretions have left the body, but a certain number grow in the very substance of the internal tissues and organs into which the germs must have previously penetrated. All these vegetable parasites belong to the class *Cryptogamia.* A few of the most important only will be briefly referred to.

During the last few years the notion that such organisms are the *causes of* disease has been very generally accepted, but like many other very popular doctrines this one rests upon very insufficient foundation. It is doubtful if many of the advocates of the fungus and bacterium germ theory of disease are aware of the multitudes of organisms like the supposed disease germs which are present in every healthy person. Wherever animal or vegetable matter (be it healthy or diseased) is undergoing disintegration and decay, low organisms grow and multiply; and I hope my readers will pause and consider before they give their assent to the doctrine that such and such a morbid condition is *caused by* the bacteria or microscopic fungi they may discover upon microscopical examination. The facts of the case may render it probable that the true cause of the pathological change had been in operation some time before the growth and multiplication of the fungi commenced.

Fig. 8.

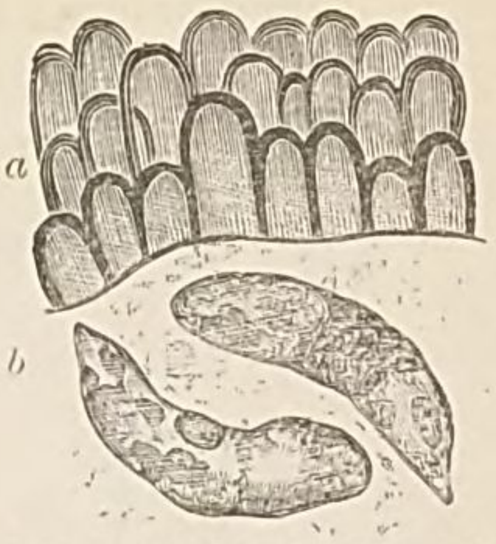


Lower or rounded extremity of one of the worm like bodies, showing how closely it is surrounded by the sarcous tissue of the muscle. × 215

Fig. 9.

Cilia-like processes, covering whole surface of envelope of worm-like body. × 700.

Fig. 10.



a, portion of membrane with hair-like processes distended with fluid and subjected to strong pressure.
b, two of the small bodies in their fully developed state, from the interior of a very long worm like cyst.
× 1800.

Fig. 11.

Cilia-like processes, covering the whole surface of the external investment of the worm-like body. × 1000.

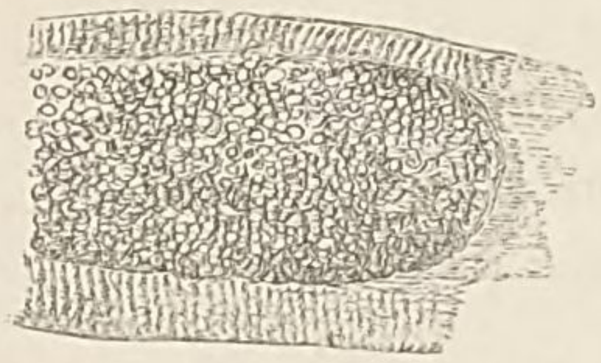
Fig. 12.

Very young worm-like body in elementary muscular fibre of the leg × 215.

Fig. 13.

Very young worm like body, from a muscular fibre of the heart of a cow. Cattle plague. × 700. *a*, muscle nucleus.

Fig. 14.



Portion of a young worm-like body embedded in elementary muscular fibre, which has been broken transversely. × 215.

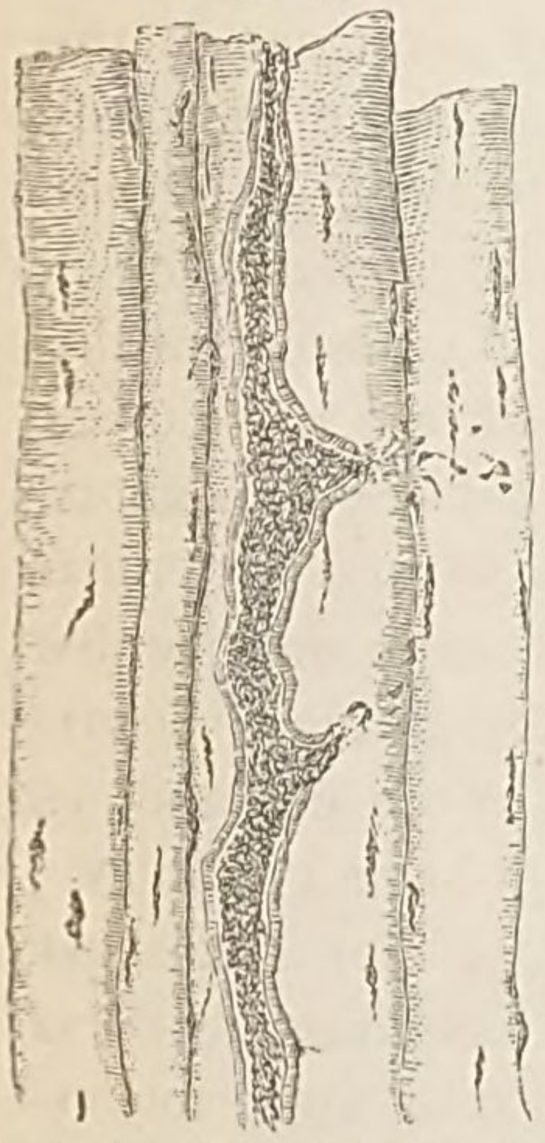
Fig. 15.

Very young worm-like body undergoing development in an elementary muscular fibre. Heart of cow. Cattle plague × 700

Fig. 16.

Tubular space in the central part of an elementary muscular fibre extending from one extremity of a young worm-like body, showing an early stage of development of the bodies found within the envelope. × 800 I regard the particles marked *a* as a young stage of the bodies represented in Fig 10 at *b*.

Fig. 17.

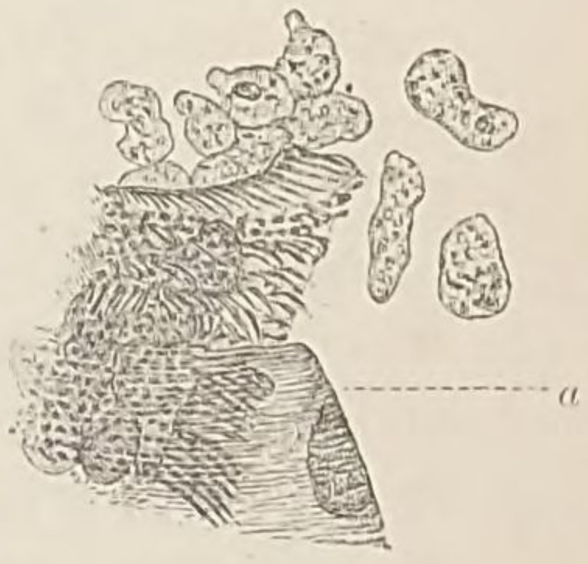


Worm like body from elementary muscular fibre. Showing escape of its contents through lateral pores or fissures. × 130.

Fig. 18.

Portion of upper extremity of one of the worm-like bodies, embedded in muscular fibre, showing the fissure, *a*, by which it gained access to the interior of the fibre at an early period of its development *b*, muscle nuclei. × 215.

Fig. 19.



Portion of one of the worm-like bodies, ruptured, showing structure of wall and escape of little bodies from its interior. *a*, muscular tissue, in which it is embedded × 700.

1000th of an inch × 130.
,, ,, |___| × 215.
,, ,, |_____| × 700.

[To face page 488.

448. Sarcina Ventriculi or Merismopœdia Ventriculi.—This alga (?) was originally discovered by Goodsir, in 1842, among the matters vomited by a patient. Since this period it has been found by a great many observers, and indeed, may now be looked upon as by no means uncommon. The sarcina is represented in pl. XLI, fig. 3, p. 290, and in pl. LXXXI, figs. 1, 2.

The vomited matters containing sarcina, have usually, but not invariably, very much the appearance of yeast, and fermentation proceeds for some time after they have been ejected. In vomit presenting these characters, the sarcinæ are, I believe, never absent; but they have been found in other cases and in other situations: by Lebert, in a case of cancer, accompanied with black vomiting; and by myself in a case in which there was a very abundant ejection of coffee-ground vomit for a few days before death. In this vomit the sarcinæ were very abundant, but there was no fermentation. The most minute sarcinæ I have ever seen are represented in fig. 1, pl. LXXXI.

The sarcina has been found in the urine, three times by Heller, once by Dr. Mackay, of Edinburgh, twice by Dr. Johnson, and twice by myself.* It has also been detected in the urine by Welcher, Johnston, and Begbie. In the fæces it has been met with frequently by Bennett and Hasse; it was observed by Virchow in an abscess of the lung, and once by Sir W. Jenner in the fluid of the ventricles of the brain. Zencker found sarcinæ in cavities of a lung affected with encephaloid disease, and also in the stomach of the same patient. He considers that it was drawn into the lung after vomiting (Henle's Zeitschrift, Band ii, Heft I). Sarcinæ have been detected in the fluid of hydrocele by Dr. Lowe, Edin. Med. Phil. Mag. N. S., July 1840; in gangrenous intestines by Demme; in cholera stools by Wedl, Mic. Journal, vol. viii, p. 163; in stagnant water by Dr. Lowe, Gardener's Chronicle, 1857; in Tinea tonsurans by Tilbury Fox; in the stomach of a rabbit by Virchow, and in that of an ape by Eberth, who also found sarcina in the cœcum of a fowl and in a tortoise.

Schlossberger considered that the sarcina was only disintegrated muscular fibre. A moderately good glass, however, will convince any one that its structure is very different from that of muscle. Dr. John Lowe agrees with Mr. Berkeley in the opinion that the sarcina is only a peculiar form of a very common microscopic fungus. Dr. Brinton and Dr. Tilbury Fox regard the sarcina as a modification of Penicillium, and therefore an ordinary mould. Dr. Brinton states that he has seen the development of the penicillium from masses of sarcina, but as spores of the latter fungus are commonly present amongst the sarcinæ, it seems possible that the penicillium might have been developed from

* For the opportunity of observing the sarcina in one of these cases, I have to thank my friend Mr. Brown, of Lichfield.

these. With these views concerning the nature of the sarcina I cannot agree. The evidence advanced in their favour appears to me wholly inconclusive. I have now carefully and repeatedly examined a great many specimens of sarcina, and have compared them with other organisms, but I am convinced that the sarcina is distinct from all other fungi, and quite peculiar.

Various plans of treatment have been employed to prevent the development of sarcinæ, but hitherto with very imperfect success. Hyposulphite of soda has been found advantageous in some cases, but the disease was not cured. Great relief to the burning sensation which frequently occurs in these cases, is experienced by the use of large doses of common salt. Several cases of this disease, with remarks, will be found reported in Dr. Todd's clinical lectures. In all the cases which have come under my own observation, the matter in which the sarcina was present was acid, although in several instances, in consequence of the ejection of much clear fluid (pyrosis), the vomit generally, had an alkaline reaction. But the brown flocculi which contained the sarcinæ were intensely acid. The sarcina is generally, but not invariably, accompanied by a great number of oval torulæ, which vary considerably in size and form in different cases, pl. LXI, fig. 3, pl. LXXXI, figs. 1, 2. These torulæ were not present in the specimens of urine which contained the sarcinæ.

By the action of acids and alkalies the sarcina becomes paler, but is not destroyed by these reagents even if warm. The cells, however, exhibit a tendency to separate from each other in a quadruplicate manner. Iodine communicates a slightly brown colour to it. It is not destroyed by the decomposition of the vomited matters in which it was developed; but in one case, in which it was present in the urine, the cells were completely broken down, and all traces of them lost, as the fluid decomposed and became alkaline. The development of the sarcina has been investigated by Frerichs in a dog with a fistula in his stomach. See also a paper "On Sarcina Ventriculi," by Dr. John Lowe, Edinburgh Philosophical Journal, new series, July, 1860.

449. Other Forms of Fungi are found in different situations; for instance, in the cavity of the mouth, especially towards the back of the tongue mixed with, and adhering to, or growing from, the cells of epithelium, will be seen, with a power of 200 or higher, a vast number of little hair-like bodies, which consist of filaments of a very minute alga (*Leptothrix buccalis*). The filaments grow upon any small particles of food which may remain entangled in the epithelium of the mouth. The papillæ at the back of the tongue are thickly covered with very long filaments, consisting almost entirely of this alga, pl. LXXXI, fig. 3; it is very abundant between the teeth, and the so-called tartar is partly composed of it. The old epithelial cells upon the tongue

and buccal mucous membrane are invaded by numerous sporules, which give to them a granular appearance under low magnifying powers, but by the aid of the $\frac{1}{1\,2}$ and the $\frac{1}{2\,5}$ the nature of the particles is readily determined. The bacteria, met with in urine and other fluids, are probably closely allied to this vegetable growth.

Similar vegetable organisms have been found in the stomach, intestines, and fæces, and in the discharge from wounds. One species occurs in the mucus of the uterus. Helmbrecht and Hannover have described minute vegetable growths in the humours of the eye.* Dr. Arthur Farre has described an alga which was passed from the intestinal canal of the genus Oscillaria.†

Dr. Tilbury Fox believes that the leptothrix and the *algal* forms of cryptogams found upon the mucus surface, are nothing more nor less than modified phases of Oïdium; 'Leptothrix' being often seen in watching the development of the 'nuclei' of the torula cells.

Dr. L. Mayer‡ found some peculiar fungus growths consisting of leptothrix with oïdium albicans over inner surface of labia, nymphæ, clitoris, vagina and vaginal portion of uterus in a case of intense itching of the vulva and neighbouring parts. The spots were visible and varied in size from a pin's head to the most minute visible point, they were bright yellow, roundish, or irregular, and were loosely attached to the mucous membrane. They rarely had the appearance of diphtheritic membranes, and were found to leave on removal shallow ulcers. The tissue on which the fungi grew was always hyperæmic, and gave out increased secretion. The secretion had a mucous opalescent, milky or creamy character, but was sometimes of firmer consistence like potato paste. In all the cases except one "the fungi were associated with more or less severe inflammation of the genital mucous membrane, and all the patients complained as soon as the mouldiness had taken root of intense burning, itching, and pricking in the vulva and vagina occurring paroxysmally and destroying rest and sleep." The troublesome symptoms disappeared with the removal of the fungi.

Many of these lower vegetable organisms require for their examination very high powers, and it is necessary to place only a small portion under the thin glass. Glycerine is a very favourable medium for the examination of fungi. The glass cover should be as thin as possible, for often their characters are not very clearly made out, unless a twelfth or sixteenth object-glass be employed. Sarcinæ may be removed with

* Quoted in Küchenmeister's "Animal and Vegetable Parasites," translated for the Sydenham Society, by Dr. Lankester, vol. ii, page 135.

† "Transactions of Microscopical Society," vol. i, page 92, old series.

‡ Dr. L. Mayer "On the Vegetable Parasites of the female genital organs in relation to practice," Monatsch. f. Geburtsk., July, 1862, quoted in British and For. Med. Chir. Rev., Oct., 1862, p. 551.

a pipette from fluids in which they subside as a deposit, or, in cases where the mass is very viscid, with the handle of a knife. If necessary, a little water may be added, and the whole covered with thin glass, which often requires to be pressed down firmly, in order to obtain a sufficiently thin stratum for examination.

To examine the so-called algæ from the mouth, it is only necessary to scrape the upper surface of the tongue, and place the epithelium and débris removed in the usual way, upon a glass slide moistened with a little water, but if it is desired to make a very minute examination of the structures, or to study the changes occurring during development under the highest power, the specimen should be well soaked in glycerine, the strength of which should be gradually increased.

450. Penicillium Glaucum.—The drawings in fig. 7, pl. LXXXI, show the general characters of this fungus, which is often developed in acid urine. It is also found in vomit, in the contents of the intestinal canal in certain cases, and in other situations. Several fungi which have been regarded as distinct species, are probably only modified forms of the yeast fungus and pencillium.

451. The Achorion Schœnleini usually appears as elongated vesicles, of a more or less oval form, figs. 1, 2, 3, pl. LXXXII, many of them being rather irregular and varying much in size, but often joined end to end so as to form branches. This fungus grows in the hair follicle, and is also found in abundance amongst the epithelium in the neighbourhood. It may frequently be seen within the hair in considerable quantity, fig. 3, pl. LXXXII, and may be found in abundance in the little honeycomb-like masses, termed favus crusts. The *favus* consists of a little cavity filled with spores of the fungus, granules, and epithelial cells, pl. LXXXV, fig. 4. One or two hairs usually pass through the centre of the favus. The fungus is composed of the *mycelium* (*a*), or the proper substance of the plant; of a *receptacle* (*b*), or *sporangium* which contains the reproductive organs; and the reproductive organs themselves, or the *spores*.

This fungus occurs in *Tinea favosa*, *Porrigo favosa*, *scutulata*, &c. The favus must be placed upon a glass slide, moistened with water, and subjected to microscopical examination. When the hair is to be examined, the same course is pursued, but it will often be found advantageous to treat it with a drop of solution of potash, which renders the hair more transparent, and the fungus more distinct. I have preserved excellent specimens of this fungus in glycerine for some years, and there is every probability of their keeping permanently.

There can be no doubt that the disease is spread by the transference of the spores of the fungus, but as in the case of many other contagious diseases, some persons escape although placed under the very same external conditions as those who are attacked. It is probable that the

Fig. 1.

Sarcinæ ventriculi, ordinary size, from vomit. *a*, sarcinæ. *b*, starch granules partially dissolved and rendered transparent. *c*, minute oval fungi usually present in vomit containing sarcinæ *d*, vibriones. *e*, oil globules. *f*, starch globule from bread, cracked, but not as yet softened. × 215. p. 489.

Fig. 2.

× 700. × 1700.

Sarcinæ, from vomit. 'Archives,' vol. II., p. 489.

Fig. 3.

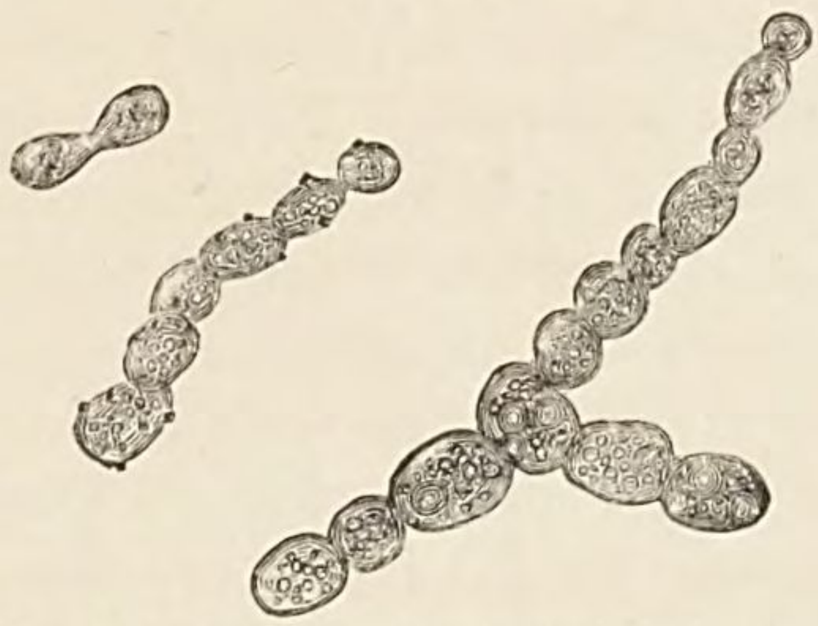
a 103

Leptothrix buccalis, from the back of the tongue and tartar of the teeth. *a*, epithelial cells, upon and in which the fungus is growing. *b*, collection of fungi. × 403. p. 490.

Fig. 4.

Penicillium glaucum × 215.

Fig. 5.

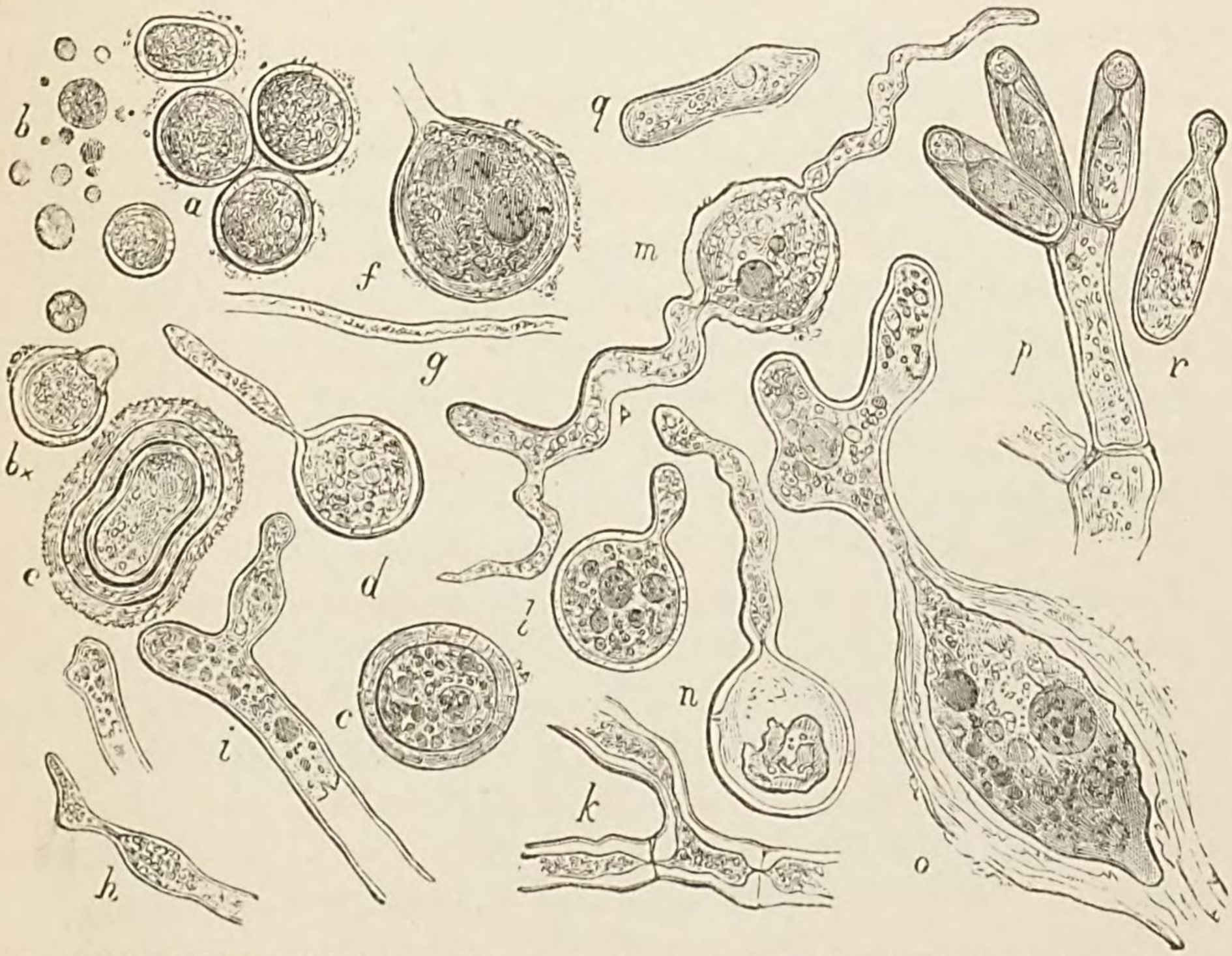


Yeast fungus from urine, covered with minute crystals of oxalate of lime. × 700.

Fig. 6.

Penicillium glaucum. × 215.

Fig. 7.



Various stages of growth of ordinary mildew. *a*, aerial spores. *b*, smallest germinal particles within these. *b*×, a spore bursting, bioplasm escaping. *c*, a spore enlarged by growth. *d*, a spore sprouting. *e*, an old spore, the formed material of which has much increased. The remaining figures show the mode of growth of the mycelium, and the fructification of the fungus, *p*, *q*, *r*. p. 492 (1859.)

1000th of an inch |—| × 215.

" " |———| × 700.

" " |———————| × 1800.

[To face page 492.

hair of those invaded is weak and its tissue imperfectly matured, and that in this way the greater susceptibility of some subjects is to be accounted for. For the disease to be successfully inoculated into healthy texture, the sporules must be kept long in contact with the epithelium. Having once commenced to grow, however, it soon spreads unless means be taken to prevent its growth and multiplication.

452. Trichophyton Tonsurans and allied Forms.—This fungus is found in the form of very minute oval or rounded, and perfectly transparent cells, *within* the bulb, and in the central canal of the hair. Its presence depends upon the hair having been broken, and the escape of the contents. It is always developed in the root of the hair. Dr. Tilbury Fox has recently shown that the earliest trace of this fungus is found at the upper part of the hair follicle. By the aid of the microscope he has detected the germs travelling towards and entering the root. It is carried onwards by the subsequent growth of the hair, and by its rapid increase, the structure of the hair is much altered; its tissue becomes dry and brittle, its fibres are split up by the growing fungus, which subsequently invades the whole of the shaft, the root, the epithelial lining, and the follicle itself, pl. LXXXII, fig. 4.

Trichophyton Sporuloides is the fungus of *plica polonica*, and closely resembles trichophyton tonsurans. It is probably a modified form of the same fungus.

Microsporon Mentagraphytes is very like trichophyton tonsurans. It is said to surround the hair within the follicle, and not to appear in the substance of the hair or outside the follicle. Dr. Tilbury Fox has, however, proved that it sometimes invades the structure of the hair, and also attacks the extra follicular part of the shaft.

Microsporon Audouini is the fungus found in *porrigo decalvans*, and is characterised principally by the small size of its spores, by the mycelium containing no granules in its interior, and by forming a tube round the hair *outside* the follicle.

Microsporon Furfur. That condition of the skin termed *pityriasis versicolor* depends upon the middle portion of the epidermis in the coloured situations being infested with the spores and mycelium of the *microsporon furfur*, fig. 5, pl. LXXXII. Cases have occurred where a previously healthy individual has been infected with the disease after having slept with a person suffering from this affection. The mycelium is wavy and branched, and the spores form characteristic groups, as in the case of the oïdium. Indeed, Dr. Tilbury Fox has adduced evidence to show that pityriasis versicolor may be produced by transplantation of the oïdium.

Puccinia Favi is found in *tinea favosa*, *tinea tarsi*, *pityriasis versicolor*, and *acne*. It is of reddish brown colour, and consists of a body and stalk. The body is composed of two somewhat conical cells articulated

to each other by their bases. The stalk joins the apex of the lower cell and the apex of the upper cell is rounded off. The stalk is flat and sometimes appears twisted. The cells may contain sporules. Dr. Tilbury Fox denies that any true puccinia occurs on the human body. Puccinia is a late stage of the ordinary uredo. That which has been called puccinia in the human subject is nothing more than a clavate terminal mycelial thread, jointed after the manner of the true puccinia, but in reality a form of penicillium. See pl. LXXXIII, fig. 2.

453. Tinea.—Dr. Tilbury Fox has proposed the adoption of the generic term *tinea* to designate the group of vegetable parasitic diseases. The diseases, with their characteristic fungi, according to this view, would be arranged as follows :—

Tinea favosa.	Syn.	Favus.	Fungus	Achorion Schönleinii.
Tinea tonsurans.	,,	Herpes tonsurans.	,,	Trichophyton tonsurans.
Tinea circinata.	,,	Herpes circinatus.	,,	Trichophyton tonsurans.
Tinea decalvans.	,,	Area.	,,	Microsporon Audouini.
Tinea sycosis.	,,	Mentagra.	,,	Microsporon mentagraphytes.
Tinea versicolor.	,,	Chloasma.	,,	Microsporon furfur.
Tinea tarsi.	,,	Ophthalmia tarsi.	,,	Trichophyton tonsurans.
Tinea Polonica.	,,	Plica Polonica.	,,	Trichophyton sporuloides.

Dr. Fox affirms that nothing but the growth of a fungus can produce the alterations of the hairs observed in the *tineæ*, and this is their *pathognomonic* lesion. It varies in degree, but is present in every instance of fully developed disease of the skin in which vegetable parasites are present—in the least degree in *chloasma*, for here the hairs are really unimportant, and the fungus chiefly attacks the epithelium. Dr. Fox has performed a good many experiments with diseased hairs out of the body, and states that he has succeeded in getting a hair containing spores which germinated and actually produced the splitting up of the hair, and other changes that are observed in ringworm, in fact "*he produced the lesion of ringworm out of the body.*"

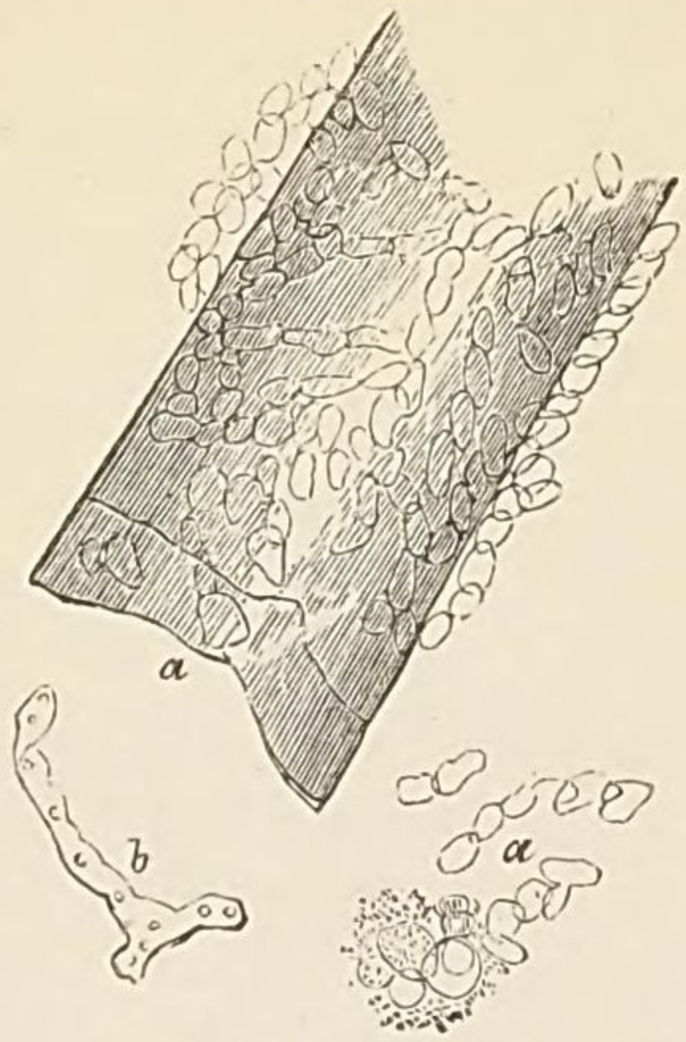
Early stage of Fungus in Tineæ.—Many observers state that the earliest trace of disease in tinea is to be observed just within the orifice of the hair follicle in an increase of the nuclei of the epithelial cells, but according to Dr. Fox's observations, this minute nuclear material is really the stroma or earliest condition of the parasite, a stage which has been overlooked by most observers. It is best seen in favus. Hence the presence of the nuclear phase of a fungus is to be regarded as grave in a prognostic sense, for a patient cannot be pronounced cured until all the minute particles are destroyed or removed, although none of the more ordinary elements of the fungus may be discovered by ordinary microscopical observations.

454. Oidium: Aphthæ: Muguet.—The aphthæ which occur upon the mucous membrane of the mouth and pharynx of ill-nourished

Fig. 1.

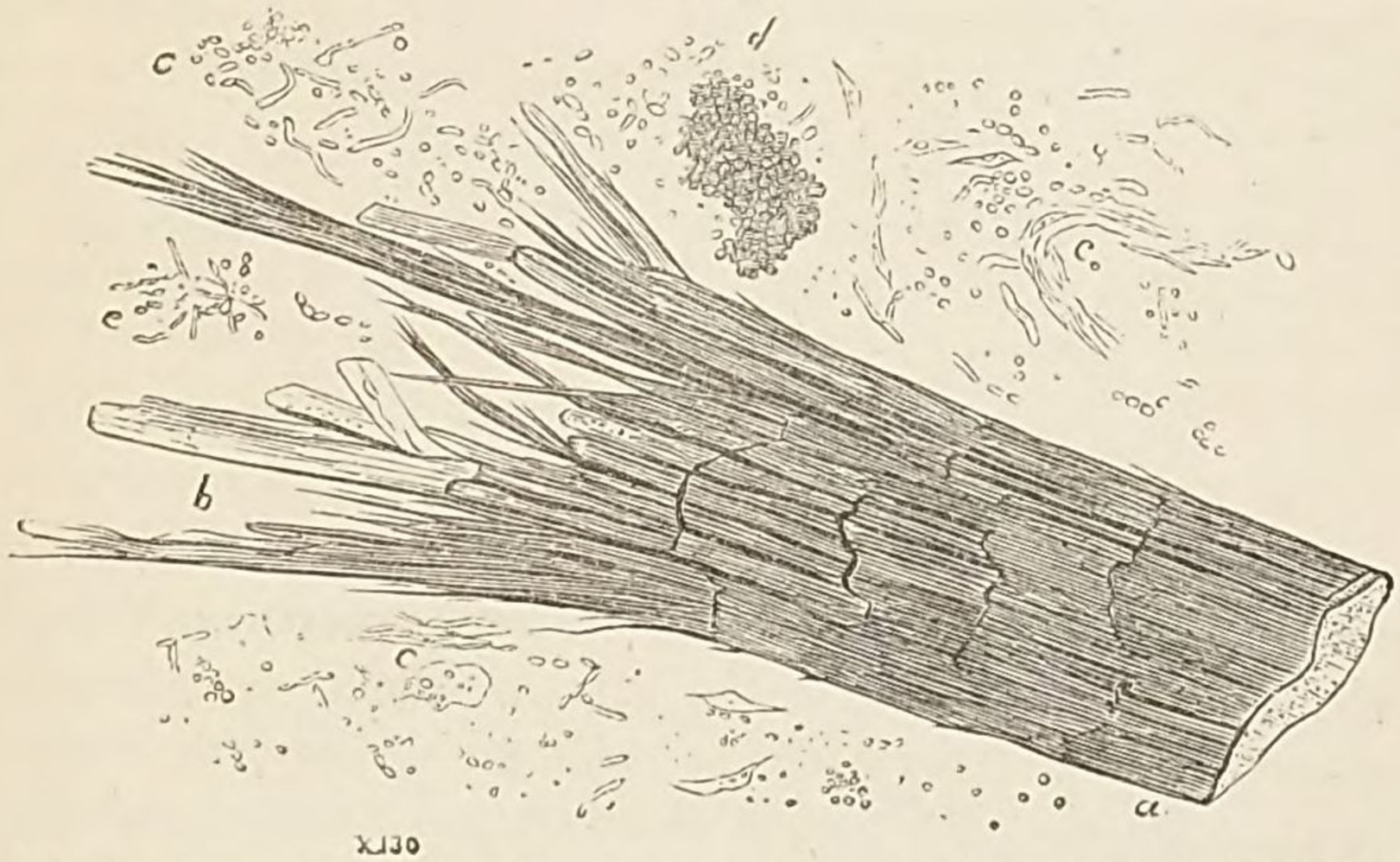
Achorion Schœnleinii, from the same specimen as Fig. 3 × 403. p. 492.

Fig. 2.



Achorion Schœnleinii. After Robin. *a*, mycelium *b*, receptacle containing spores. p. 492.

Fig. 3.



×130

Achorion Schœnleinii in various stages of growth, from the crusts of a very bad case of Porrigo favosa. The boy's scalp was completely covered It had been entirely neglected, and was of eleven years duration. The hair was everywhere brittle, and its fibres readily separated from each other. *a*, hair broken near the root. *b*, extremity showing separated fibres *c*, altered cells of squamous epithelium from the skin. *d*, sporules of the fungus, forming a dense collection. *e*, sporules and thalli of fungus separate. × 130. p. 492.

Fig. 4

Root of hair invaded by the spores of trichophyton (tinea) tonsurans. After Tilbury Fox. p. 493

Fig. 5.

Microsporon furfurans, or fungus of tinea versicolor (chloasma). After Tilbury Fox. p. 493.

[To face page 494

infants, and the whitish matter resembling false membrane, which is sometimes found in the same situations in adults, who have long suffered from exhausting diseases, and to which the term *muguet* has been applied, are composed of a vegetable fungus, which was first described in 1842, by Gruby, and has been spoken of by him under the names of *aphthaphyte* and *cryptogames du muguet*. It is placed under the genus Oïdium, and termed *Oidium albicans* by Robin.* The appearance of this fungus is shown in fig. 5, pl. LXXXIII, fig. 1, pl. LXXXIV. It is also found in vomited matter.

455. Diphtheria has been considered by some observers to be intimately connected with the development of a particular vegetable growth. Thus the contagious character of the disease has been accounted for. Where, however, the fungus is found, its presence may be due to other circumstances. The false membrane is no doubt a nidus very favourable for the development and growth of fungi. It is quite certain that in many cases of diphtheria, among which may be reckoned those which have fallen under my own notice, there was no vegetable growth to be detected in the false membrane removed from the fauces.† The microscopical characters of the false membrane are described in page 284.

456. Fungus found in the External Meatus of the Ear. Aspergillus?—The vegetable growth represented in pl. LXXXV, fig. 3, was removed by Dr. Grove from the external meatus of a gentleman in good health, who had been suffering from inflammation of the canal. The specimen was given by Mr. Deane to Dr. Sturt, who kindly allowed me to have the accompanying drawing of it made.‡ A case in which a fungus, of the same kind in all probability, was found in the external meatus of a girl, aged eight, is given by Mayer. She was a scrofulous child, suffering from discharge from the ear. Many filaments contained a receptacle filled with spores.§ Link considers this fungus to be a species of Aspergillus, and Robin places it in the same genus.‖ A species of Aspergillus has been detected in the human lung by Prof. Dr. Carlos May Figueira, of the Medical School of Lisbon, ("Jornal da Sociedade das Sciencias Medicas de Lisboa," No. 10, Outubro de 1862).

457. Psorosperms and Gregariniform Bodies.—Hair is sometimes the seat of growth of a very curious organism belonging to the psoro-

* "Histoire Naturelle des Végétaux Parasites qui croissent sur l'homme, et sur les Animaux vivants," Paris, 1853. See also a review of this work, by Dr. Parkes, in the "British and Foreign Medico-Chirurgical Review," October, 1853.

† In one case there was some algæ, but it was afterwards proved satisfactorily that these had been introduced after the removal of the false membrane from the patient's mouth.

‡ The case, accompanied with a drawing, is given in the "Transactions of the Microscopical Society," new series, vol. v, page 161.

§ Müller's "Archiv," 1844, page 404.

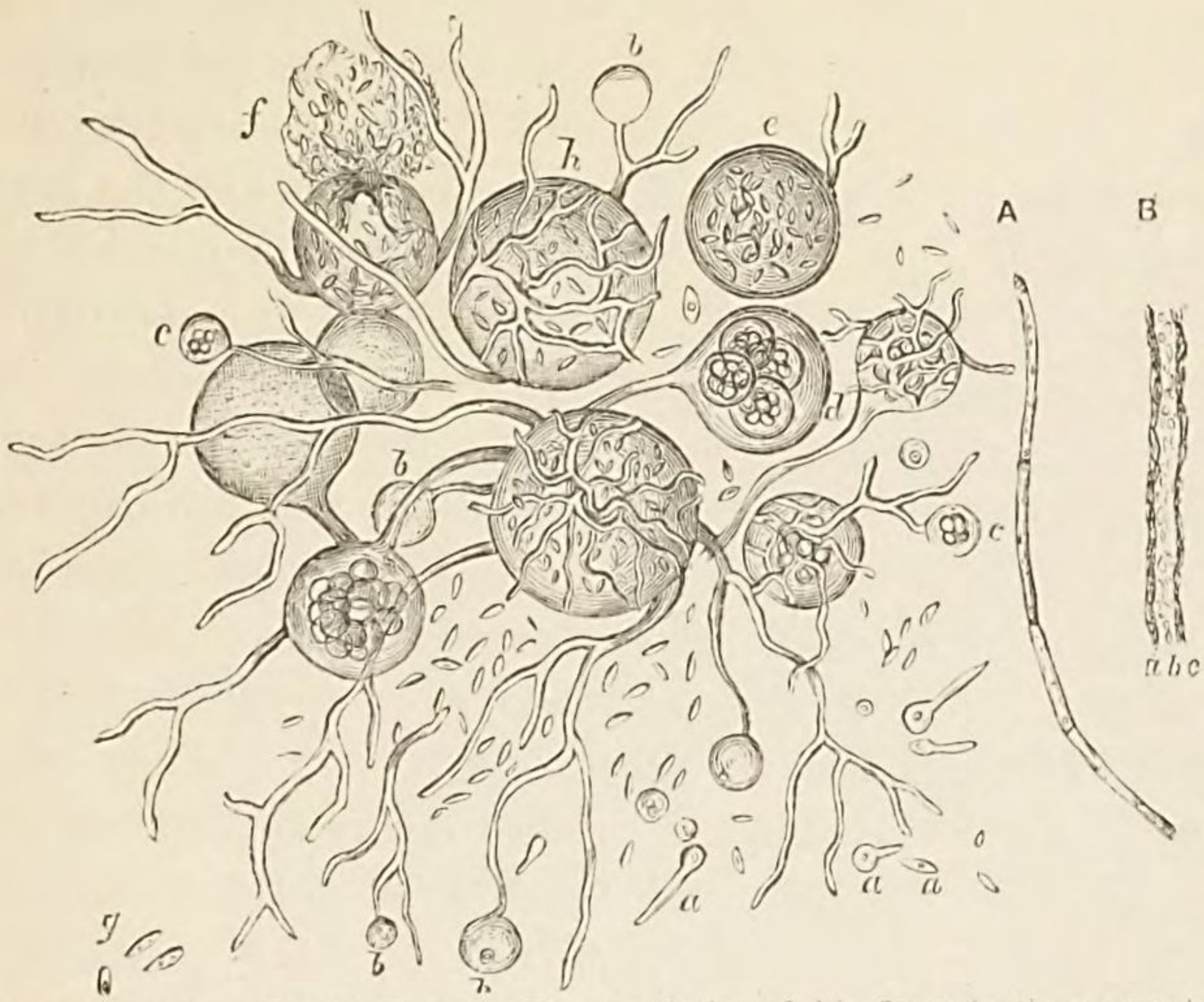
‖ "Histoire Naturelle des Végétaux Parasites," par Ch. Robin, 1853.

spermiæ, of the exact nature of which we know very little. Psorosperms are found in all the vertebrata, and in connection with many of the lower forms. Organisms of this class constitute the disease known as pébrine, which affects silkworms, and which has been carefully studied by Pasteur, who considers that psorosperms belong to the vegetable kingdom, and who has proved that these bodies contain a substance nearly allied to cellulose. These bodies are extremely minute, and a magnifying power of from 500 to 1000 diameters is required for investigating them. Lindemann discovered them upon the hair of a chlorotic unhealthy girl. Many of the hairs had psorosperms attached to them, but upon some moving gregarinæ were found (quoted by Leuckart, vol. 1, p. 741). Psorosperms have been discovered in the liver by Dressler of Prague, and also by Virchow, and in the human kidney by Lindemann of Nishni Novgorod. They are common amongst fishes. The simplest of the remarkable bodies described in the muscles of the pig (see p. 486) seem to be of this nature, but the very curious integument or envelope with its bristle-like surface seems too complex in its structure to belong to an organism so low in the scale as psorosperms are supposed to be.

458. Chionyphe Carteri is the fungus discovered by Dr. H. V. Carter in the fungous foot disease of India. According to Dr. Carter, there appear to be three principal varieties of the disease and two principal varieties of the fungi. In the first the fungus occurs in globular masses, sometimes as large as a pistol bullet, black externally, brown within, and having a radiated appearance on section. The radiated fibres end in globular expansions, rendering the exterior surface tuberculated, and consist of cellular threads, branching and anastomosing. Interspersed here and there are a few granules and large oval cells placed end to end. The globular expansions are composed of fibres formed of large oval cells, united at their ends with oval nucleated spores, situated between the fibres here and there. These globular expansions often become detached, and occur as small granular masses. In the second variety the fungus is always in the form of small particles. These may be light coloured and composed of threads formed of round or oval cells, mixed with granular matter, granular cells, and oil globules, or they may be red or pink grains visible to the naked eye, made up of minute beaded fibres or nuclei, sometimes single and then oval in form, sometimes double, triple, or quadruple, and then angular. The fungus particles may also consist of light brownish granules, made up of minute bodies, each of which has a crystalline fatty envelope, or of similar granules, whose structure is that of the black fungus.

Dr. Carter's drawing has been copied in pl. LXXXIII, fig. 1, reduced to one-fourth the size. The illustration represents a specimen of the *red fungus*, which grows on the surface of the fluid covering the

Fig. 1.



Chionyphe Carteri. After Dr. CARTER. The description of this figure is given on page 497. × about 250 (reduced from large drawing). p 496

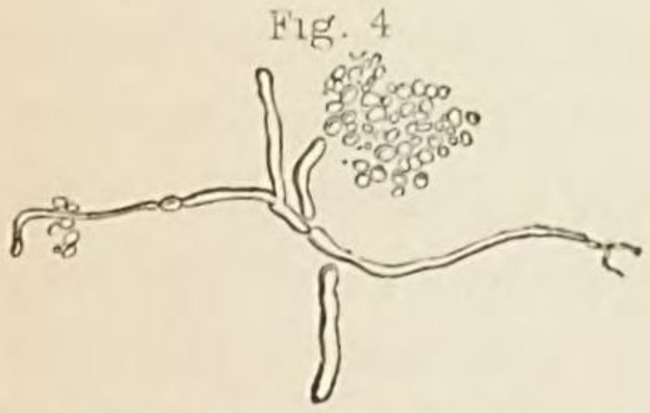
Fig 2.

Penicillium, showing development in form of TORULA, *a*; and in that of PUCCINIA, *b*. After TILBURY FOX. p. 494.

Fig. 3.

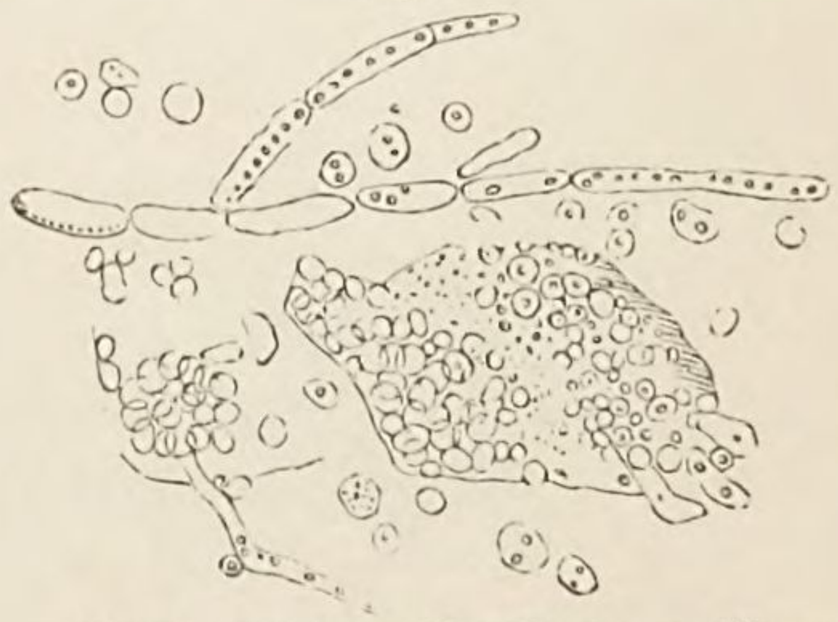
Mycelial form of fungus. Tinea circinata (herpes circinatus). After TILBURY FOX.

Fig. 4



Fungus formed in tinea tarsi After TILBURY FOX.

Fig. 5.



Oidium albicaus. After ROBIN. p. 495

1000th of an inch ⟨——⟩ × 250.

[To face page 496.

portions of a foot affected with the "Black Fungus,"—magnified to show its development from the germinating sporidia *a*, *a*, *a*, to the formation and bursting of the spore *f*. *a*, *a*, *a*, *a*, germinating sporidia; *b*, *b*, *b*, commencement of spore-cells containing nucleus; *c*, nucleus and contents of spore-cell further advanced; *d*, apparent quadruplication of contents of spore-cell, with further sub-duplication of their interior; *e*, spore and sporidia formed; *f*, spore bursting; *g*, sporidia more highly magnified, to show shape and nucleus; *h*, spore embraced by a condensation of small filaments—very common if not constant. In the figures under A, a filament is represented which is composed of cells with a nucleus at one end of each, and under B, is represented the 'felt-like' form of the layer of red fungus as it grows in the bottle; *a*, filamentous layer; *b*, layer of spores; *c*, filamentous layer below.

458. Other Forms of Fungi.—Low forms of cryptogamia have also been found in the lung by Professor Bennett, and have been noticed in the stools by him and other observers. Meissner describes a fungus which he found amongst the cells of the nails of an octogenarian. The nail was rendered transparent by caustic soda. It was permeated in every part by the fungus. Fungi have been found in glandered lungs; on the pleura (Roger and Gardner); in the expectoration of phthisis (Remak); and in the kidney by Tonge and Powell, pl. LXXXV, figs. 9, 10.

There is a group—*Leptomitus*, whose "characteristic feature is an enlarged ovoid cell, mostly terminal with a little projecting joint at the apex, and containing more or less nuclei—the whole resembling a club very much" (Fox). The following varieties have been found, *L. urophilus* in the urine; *L. Hannoveri* in the œsophagus and stomach; in typhus by Robin; in bronchitis by Fuchs. *L. uteri*, called by Dr. Wilkinson, Lancet, 1849, p. 449, *Lorum uteri*, in consequence of the supposed breaking up of the terminal cell into several threads, so as to resemble a rash, and another form has been detected by Gubler in the epidermis in a case of gun-shot wound of the arm. Dr. Tilbury Fox has clearly shown that all the forms of Leptomitus are forms of oïdium. The terminal and aerial filaments bearing the oïdial, and the basal the leptomitus character, see pl. LXXXIV. The Lorum uteri is nothing more nor less than a free branching or budding out from the large ovoid cell of the ordinary Leptomitus.

459. The Identification of many of the fungi above referred to is a most difficult matter. Indeed, unless the organism can be studied at different periods of its development, it is almost impossible to assign to it its proper name. Moreover, the drawings given in different works when original differ so much from one another that not only students but teachers and authorities are often much perplexed. Moreover, all these fungi at an early period of development are but minute particles

which could not be distinguished from the minute sporules of bacteria which exist everywhere. Some authorities maintain that fungi referred to distinct species may be developed from the same germ particle, and that the differences in the fully developed state of the fungus are due not to inherited and transmissible specific peculiar characteristics, but to conditions present during the growth of the organism. This difficult question bids fair to become more and more involved by reason of the views now accepted upon, as I believe, erroneous data concerning the relation of bacteria to various contagious diseases.

460. Of the Mode in which Fungi Gain Entrance into the System of Living Beings.—Vegetable organisms found even in the substance of the inmost tissues result from the development of germs introduced from without. Since we know that the germs of many entozoa make their way very readily through the tissues of the organism, there is no difficulty in explaining how bodies so very much smaller than these, as the germs of the fungi, are introduced. These germs have the power of insinuating themselves through the firmest tissue and multiply in number as they make their way through. The appearance of some of the most minute living particles of simple fungi are represented in pl. LXXXV, figs. 5, 6, but there is reason to think that they possess individual powers of growth and multiplication long before they have grown large enough for us to see them even with the aid of the highest magnifying powers we possess.

Dr. Fox enumerates the modes of invasion as follows: "(1), Through natural orifices; (2), That in which the growing force forces the mycelial thread beneath the layers of the superficial tissues; (3), That in which processes shoot out from the spore and enter by openings such as stomata in plants; (4), That where the cells' contents alone are absorbed; (5), That in which the spores are carried bodily inwards by growing parts; or (6), Dissolve away the opposing structures by chemical action (as in the hard shells of molluscs); or (7), Enter by traumatic lesions as in the case of the fungus fort of India. In each and every instance the germs of the parasites are derived *ab externo*, and not generated spontaneously." Lastly it must be added that the germs of bacteria, and probably of other low organisms, may long remain in a quiescent state in the tissues and fluids of the body, ready to be developed should the conditions become favourable.

Even the vegetable nature of fungi has been called in question by some, but it would be absurd to reply to those who entertain such utterly untenable opinions. Any one who knows how to use a microscope can convince himself of the truth of the statements in the text upon this matter without any difficulty.

The part played by fungi in the production of disease has been much disputed. Some regard their presence as merely accidental,

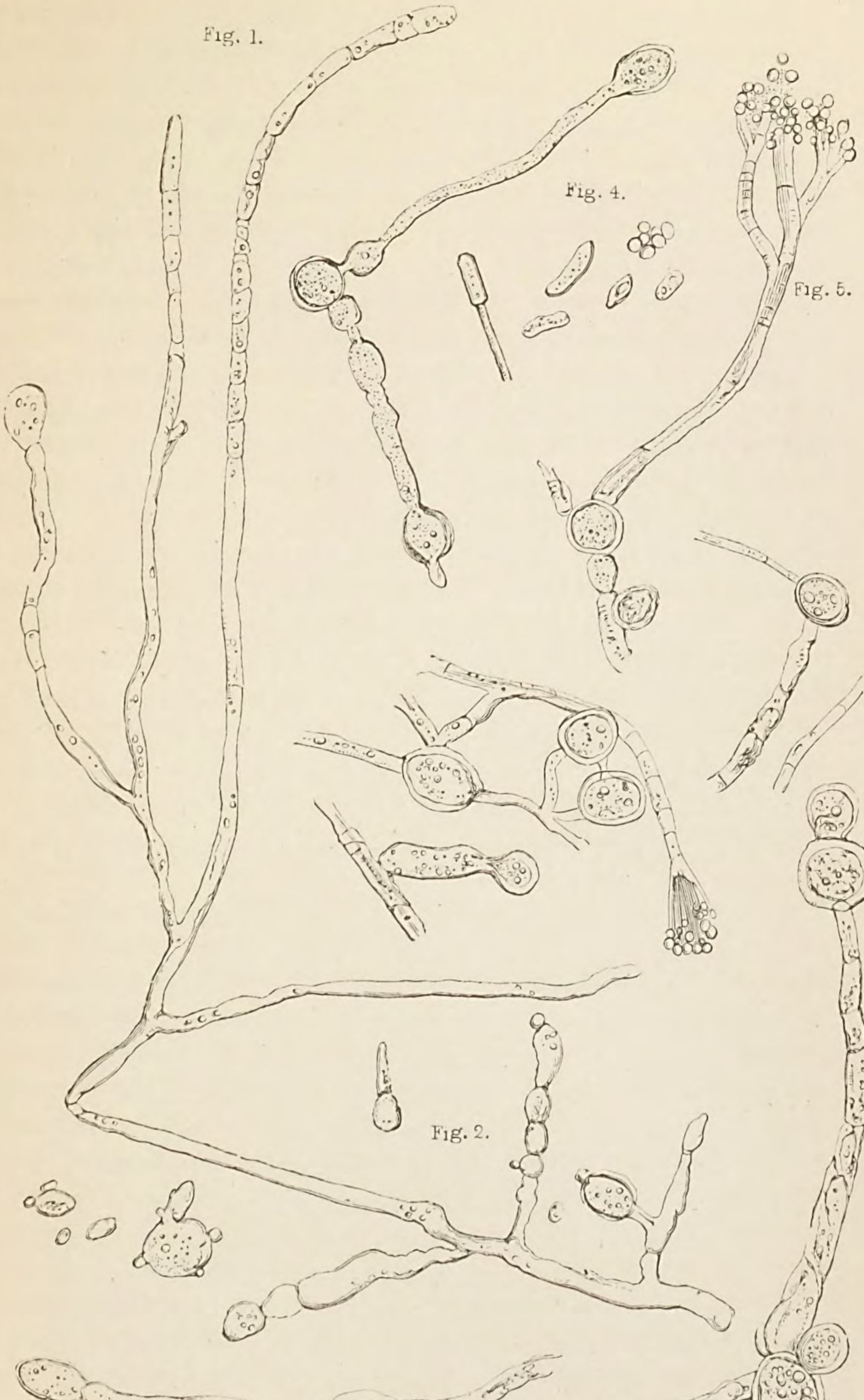


Fig. 1.

Fig. 4.

Fig. 5.

Fig. 3.

Fig. 2.

Fig. 6.

Fig. 1, Oidium albicaus Fig. 2, Leptomitus Fig. 3, Leptomitus from the germination of a torula. Fig. 4, Other forms assumed by torula Fig 5, Penicillium, showing sacculi in the mycelium. Fig. 6, Oidial threads from torula. After Dr. TILBURY FOX. p. 497.

[To face page 498

others consider them the sole cause of the diseases with which they are associated. There can be no doubt that a certain state of the tissues is necessary for the free growth of fungi. Young and actively growing tissues resist their invasion, while old textures of all organisms, vegetable as well as animal, are often attacked and partially destroyed by them. Any conditions which favour the production of an unusually soft formed material, favour invasion at an earlier period than if the tissues were firm and the growth well matured. If the formed material be produced very quickly, it is more likely to be attacked than if it is very slowly formed. This is well seen in the higher vegetable tissues. The weak and sickly plant succumbs to fungi while the strong and vigorous one remains untouched, although equally exposed to attack. The wood of the elms in and near London is rendered rotten and useless by the invasion of microscopic fungi, and this is probably to be explained by the conditions to which the plant has been exposed during its growth, and which were not favourable to the production of firm ligneous tissue. Practitioners are well aware that weak, ill-fed scrofulous children often suffer from parasitic diseases, which strong and healthy ones, although placed under precisely the same conditions, escape.

461. Examination of Vegetable Growths.—The examination of these vegetable growths in the microscope presents no difficulty; but without care they may readily be passed over unobserved, as their structure is very delicate, and they are generally accompanied with epithelial cells and much débris. A very small piece only should be submitted to examination, and should be moistened with a little water, glycerine, or dilute syrup. They may be seen with a power of 200; but to bring out their characters clearly, a power of from 500 to 800 is required. All may be preserved in glycerine. The method of preparation described in Chap. VII, page 98, is of great value in the investigation of the structure and mode of growth of the various forms of fungi, but, owing probably to the difficulty of destroying their life without completely altering their appearance, staining with carmine is difficult. They should be kept for some time at a temperature above 100 while immersed in weak glycerine, before they are placed in the carmine fluid, or should be killed by the action of alcohol. Like other tissues, most of them may be well preserved in strong glycerine, to which a little free acetic acid has been added.

The bioplasm of all the fungi may, however, be stained in the carmine fluid. It has been found necessary to modify the process somewhat in order to increase the power of the solution to permeate the external membrane which protects the living matter. About one-third of its volume of alcohol is added to the fluid, and in some instances it is necessary to expose the fungi to a temperature gradually increasing to about 160; for it has been found that the living bioplasm resists the

action of the carmine fluid. In some cases I have succeeded in staining the bioplasm satisfactorily, but after the fungi had been mounted for some months, the bioplasm began to grow, and from the red mass a colourless mycelium gradually extended itself. The vitality of the bioplasm of some fungi is most remarkable. I have many specimens which have been thoroughly well preserved in the strongest glycerine for twenty-five years which contained living sporules, and in not a few these were in a state of very slow growth—but there are reasons for believing that many of the lower fungi, at least in the sporule state of development, may retain their vitality for centuries.

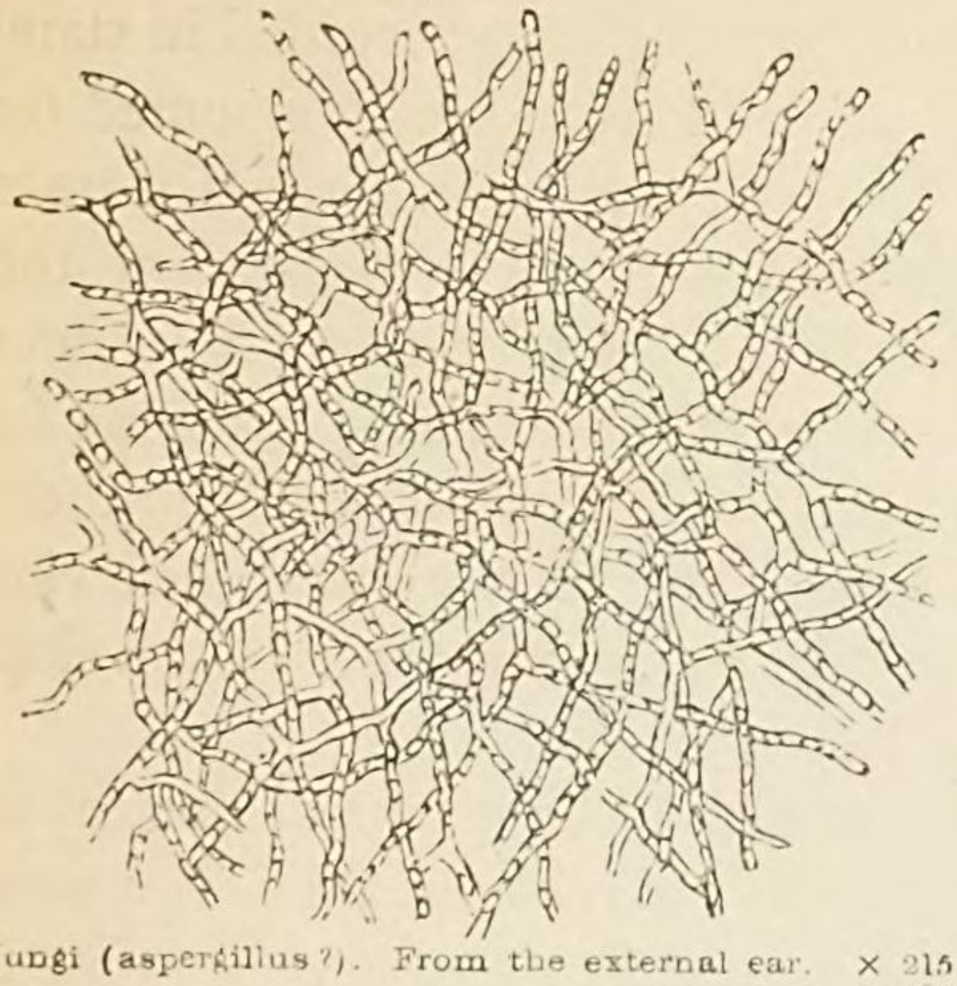
Another method of rendering the vegetable organism distinct, is to soak the tissue supposed to contain it in glycerine, but this is a slow process. More than a week's soaking is usually necessary, but time is saved by placing the specimen in a watch-glass in a few drops of a solution consisting of equal parts of glycerine and water. The watch-glass lightly covered with a piece of clean paper to keep out the dust may be put in a little hot water oven or other arrangement by which evaporation at a temperature not above 100 can be ensured, or better the watch-glass may be placed under a bell jar with a dish containing a little strong sulphuric acid.

In order to demonstrate the fungi in the elementary parts of hair, or of cuticle, as has been already stated, liquor potassæ will be found of advantage. The dry cells of the hair, although they may be full of fungus spores, show nothing if examined in water in the usual way, but if a little of the alkali be added the hard horny tissue soon becomes softened and transparent, when the little oval sporules and perhaps branching threads become quite visible. In most cases it is better to work with a magnifying power of from 300 to 1,000 diameters.

On the subjects discussed in this chapter the following works may be consulted :—

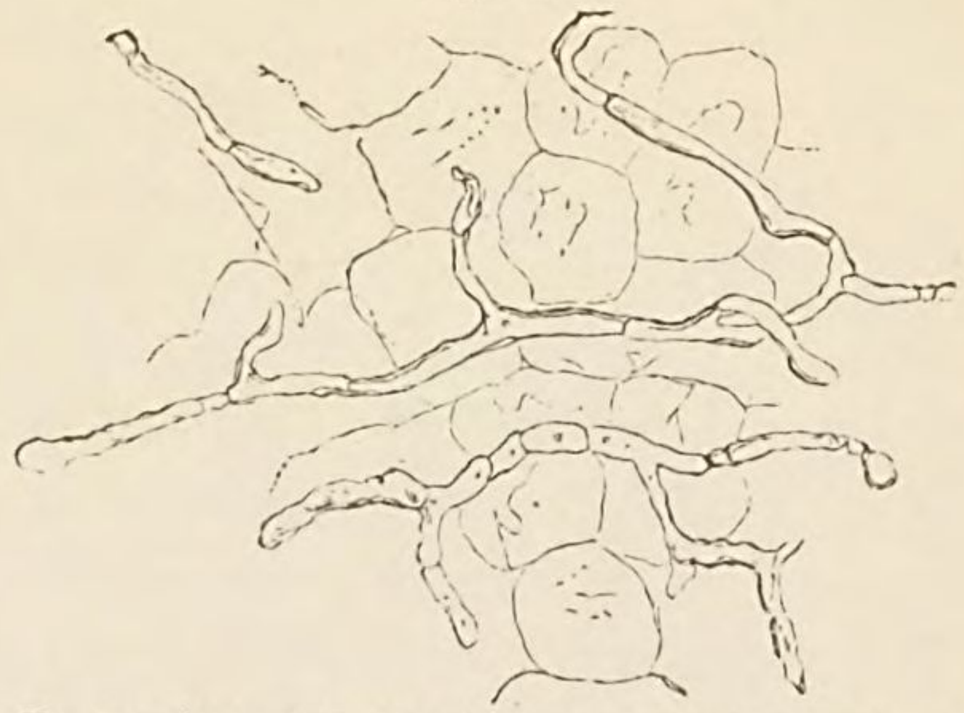
George Nayler, "A Treatise on diseases of the Skin." Cobbold, "On Entozoa," also "On Tape-worms." Dr. Althaus, "Trichina Disease." Robin, "Histoire Naturelle des Végétaux Parasites," Paris, 1853. Wedl's "Elements of Pathological Histology," translated for the Sydenham Society, by Professor Busk. Cazenave, "Annales des Maladies de la Peau et de la Syphilis." Bazin, "Recherches sur la Nature et le Traitement des Teignes," Paris, 1855. Bennett, "Transactions of the Royal Society," Edinburgh, 1842, vol. xv, and "Lectures on Clinical Medicine," 1858. Gruby, "Comptes Rend.," 1843-44. Rayer, "Traité des Maladies de la Peau," Paris, 1835. Papers in the "Transactions of the Microscopical Society." Küchenmeister's "Manual of Animal and Vegetable Parasites," translated for the Sydenham Society, by Dr. Lankester. Tilbury Fox "Skin Diseases of Parasitic Origin, including the history and relations of the fungi formed in man." Hillier, "Handbook of Skin Diseases." Dr. McCall Anderson, "The Parasitic Affections of the Skin." Dr. Squire, "Atlas of Skin Diseases."

Fig. 1



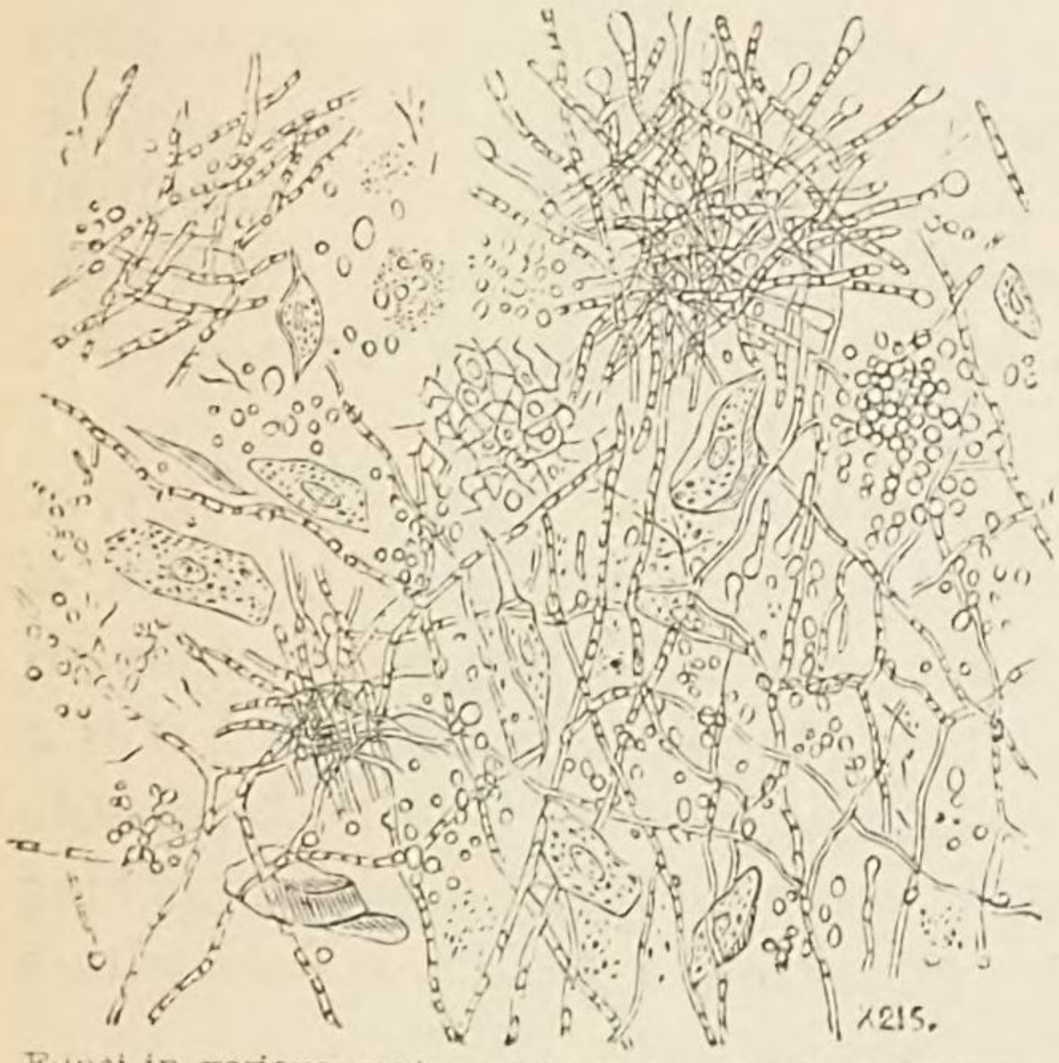
Fungi (aspergillus?). From the external ear. × 215 p. 495.

Fig. 2.



Fungus (mycelial form) among epithelial scales. Tinea circinata. After Tilbury Fox.

Fig. 3.



Fungi in various stages of growth, expectorated by a patient in the last stage of phthisis. × 215.

Fig. 4.

Portion of a favus crust, with Achorion Schœnleinii. *a*, mycelium. *b*, receptacle. *c*, spores. p 192.

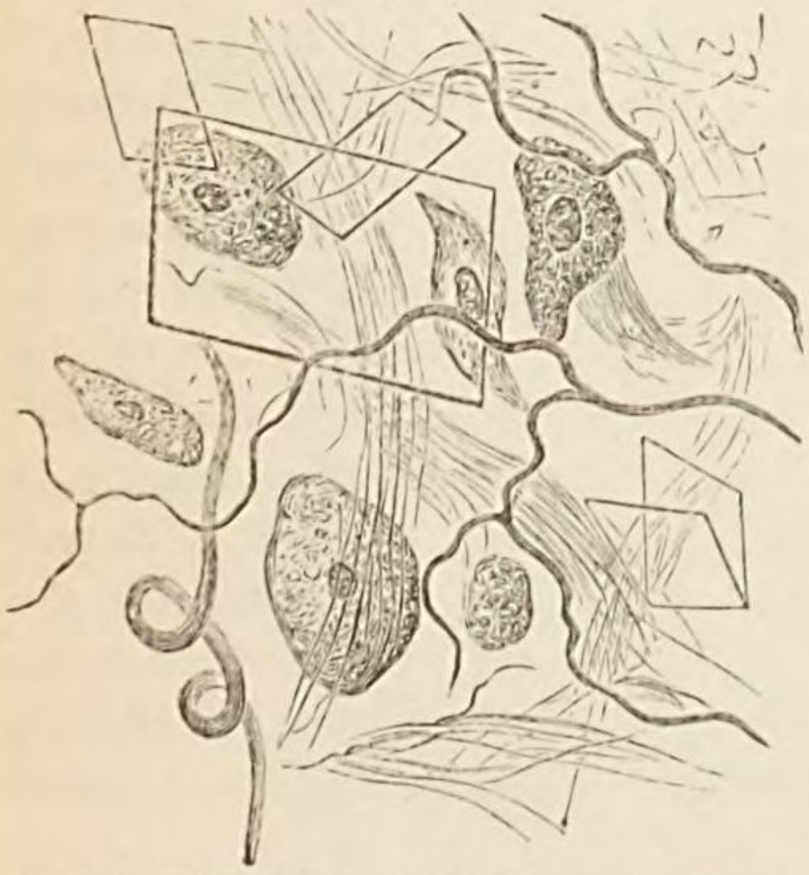
Fig. 5.

The most minute germs of fungi visible under a $\frac{1}{50}$ The smallest less than $\frac{1}{100000}$ in. in diameter p. 498.

Fig. 6.

Sporules of fungi and bacteria. × 215. p. 498.

Fig. 7.

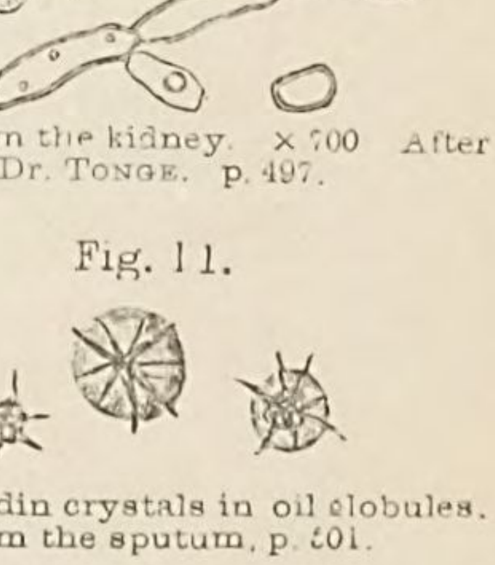


Small sub-cutaneous sebaceous tumour, about the size of a filbert, from beneath the integument of the abdomen Altered fibres of white and yellow elastic tissue, plates of cholesterine, and modified epithelial cells. × 215. p 374.

Fig. 8.

Blood from a case of leucocythemia, showing a great number of white blood corpuscles (Omitted from Plate XXXVII, p. 263.)

Fig. 9.



Fungi found in the tissue at the apex of the pyramids of the kidney. After Dr. Tonge. × 215.

Fig. 10.

Fungi from the kidney. × 700 After Dr. Tonge. p. 497.

Fig. 11.

Hæmatoidin crystals in oil globules. From the sputum, p. 501.

1000th of an inch ⌊——⌋ × 215.

" " ⌊————————⌋ × 700.

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CONCLUDING NOTES.

462. Note to Chapter on Spectrum Analysis.—Since the chapter was printed, Dr. Edward Lawton Moss has discovered a spectrum band that appears to be new in the urine of a case of cirrhosis of the liver. The line in question lies between 1,700 and 2,100 of Kirchoff's scale, and just includes the "F solar line in its right edge." The line was not due to bile but it corresponded to a line produced in "normal acid fæces," but which was absent in "healthy alkaline fæces." The line disappeared when the urine was neutralized with ammonia and re-appeared on re-acidulation. On Pathological Absorption Spectra, by Edward Lawton Moss, M.D., F.R.C.S., Surgeon, H.M.S. "Alert." Medico-Chirurgical Transactions, 1875.

463. Substances in Sputum.—In addition to the matters referred to in Chapter XIV as constituents of sputum, I omitted to mention how portions of bone have been found in sputum. A remarkable instance occurred not long ago in a case of caries of some of the dorsal vertebræ —followed by pleurisy and pneumonia. The portion of bone was as large as a pea and consisted entirely of cancellated structure. Another case is mentioned by Friedreich. The same observer also found sarcinæ in sputum as well as amyloid corpuscles. In a case of expectoration of fibrinous casts of the bronchi—Friedreich discovered crystals which he found to consist of tyrosin.

Crystals of hæmatin and hæmatoidin are not unfrequently found in sputum in cases in which blood has remained for some time in the bronchial tubes, in the air-cells or in a cavity. The coloured crystalline matter is often seen in a large oil-globule. Of this a good example has been figured in pl. LXXXV, fig. 11.

464. Mucus Casts, large Bowel.—See page 292. Sometimes these curious cords, or cylinders of modified mucus attain a large size and acquire a degree of firmness in consistence which is very remarkable. I have often seen them much firmer than the fibrin which may be taken from the heart after death, and of a dark yellow colour. The general appearance and consistence is in fact such as would render it impossible to feel sure of their nature without careful microscopical examination. I have myself mistaken portions of mucus material for pieces of white and yellow elastic tissue until by microscopical examination, the absence of definite structure was proved, while the fibrillation and the presence of numerous little granular mucus bioplasts and modified epithelial particles convinced me of the real nature of the matter under examination.

One of the most striking specimens of the kind was brought to me for examination by Dr. Eatwell. It was passed from the bowel of a

patient of 72 years of age, who discharged several such masses, generally once in a month. Some of these were two feet in length. They varied much in diameter, but in some places were as thick as the finger. Mixed with these smooth, firm, cylindrical moulds were many irregular shreds and fragments. As the whole mass passed lay in the bottle in fluid, it might have been mistaken for a half decomposed spinal cord with its nerves. It was of a yellow colour—not white in any part, nor dirty white. The patient vomited previous to the bowel attacks, and brought up sarcinæ, but these organisms had nothing to do with the formation and escape of the mucus casts from the bowel, though the presence of sarcinæ may no doubt be accepted as additional evidence of an abnormal state of the mucous membrane of the digestive tube.

465. On Preparing and Cutting Sections of ordinary Soft Tissues; e.g., Skin, Glands, Tumours, &c.—Dr. Pritchard sends me the following note: Place a small portion of thin well washed tissue in a weak chromic acid solution ($\frac{1}{3}$ to $\frac{1}{4}$ per cent.), and allow it to remain there two or three days. This slightly hardens the tissue, brings out the various delicate outlines, and prevents the softening and solvent action of glycerine, which is so apt to spoil fresh tissues or those merely hardened by alcohol. Then make sections by means of one of the freezing microtomes (page 439); wash the sections in water, transfer and preserve them in strong spirit until convenient to stain and mount.

These sections may be stained by any but the metallic staining agents, and mounted in glycerine, glycerine jelly, acetate of potash solution, Canada balsam, or damar, but the first of these yields the best results as a rule.

466. An easy method of Carmine Staining, even with Tissues hardened in Chromic Acid.—Dr. Urban Pritchard recommends that the sections or small pieces of tissue be placed in a diluted carmine solution; *i.e.*, Beale's solution 1 part, water or proof spirit 2 parts. In from 24 to 48 hours the tissue will be stained throughout, both the protoplasmic (bioplasm) and formed material. The sections may then be washed in water and transferred to a $\frac{1}{2}$ per cent. solution of hydrochloric acid in water or proof spirit. In from 1 to 3 hours the acid will dissolve out the carmine from all but the protoplasmic portions. The sections may again be washed, and then preserved in strong spirit until convenient to mount. This process is simple, and very easily carried out, as it is of little consequence how deeply the tissue may have been overstained in the first instance. It may be applied to most tissues, except brain and spinal cord, in these the acid dissolves out the carmine from the nerve cells and axis cylinder, but stains the nuclei of the neuroglia, thus resembling Ranvier's purpurine process of staining.

467. Dr. Pritchard's Microscope for examining the Circulation in the Frænum of the Tongue.—The frænum of the tongue is the part chosen for observation, and for this purpose a well developed frænum, such as is found in a so called tongue-tied person, should be selected. The microscope, pl. LXXXVI, fig. 5, p. 504, is so held that the frænum fits into the space left by the connector (F) between the speculum and tube E; the mouth may then be nearly closed, and the speculum must be directed so that the light passes directly through the whole apparatus. The length of the tube E very nearly corresponds to the focal length of the combination of lenses, and the observer, looking through in the ordinary way, focuses the object by withdrawing the eye-piece more or less as may be necessary.

The objective here figured is No. 2 Hartnack ($\frac{3}{4}$ inch), with a medium eye-piece. This combination shows the circulation perfectly; and the individual corpuscles may be distinctly made out; but should higher powers be desired, the apparatus can be readily adapted for them by making the tube E shorter.

468. Further observations on Filaria Bancrofti, by Dr. Cobbold.—In the "Lancet," for October 6th, 1877, Dr. Cobbold has published further observations upon this interesting entozoon, in continuation of his remarks in the same journal of July 14th, 1877, which I have transcribed in pp. 483, 4, of the present work. Dr. Bancroft forwarded some excellent photographs which Dr. Cobbold has shown to me. One is that of a youth with a large tumour in the axilla caused by the presence of filariæ. Dr. Bancroft names the condition *Helminthoma elastica*, and refers to cases of elephantiasis and varix, which were also produced by this entozoon.

Dr. Cobbold gives the following account of his examination of the worms received from Dr. Bancroft, on August 28th, 1877:—

"The filariæ were enclosed in four small tubes and preserved in glycerine. Three of the tubes (marked 1, 2, 3) contained sexually mature worms; the fourth being labelled 'Sediment from adult *Fil. sang.*—young and ova.' I shall describe their contents in succession.

"On the 6th of September I examined the filaria in tube No. 3. The specimen was injured and in four portions; these collectively measuring three inches in length. Although, to the naked eye, the worm appeared to Dr. Bancroft to be of the thickness of an ordinary human hair, yet actual measurement showed it to be $\frac{1}{90}''$ at the thickest part. Notwithstanding mutilation and partial evisceration, I made it out to be a female.

"At the same time I likewise examined the specimen in tube No. 1. This was also a female. Towards the centre of the body an accidental hernial protrusion of the uterine horns and intestine had taken place. In a rough lithograph sent by Dr. Bancroft, this specimen is figured

and described as the 'parent worm of the filaria sanguinis, emitting young filariæ from two loops.'

"On the 14th of September I examined the contents of tube No. 2. In it I found one tolerably perfect female filaria, and also a delicate shred forming part of one of the uterine horns of another worm. This filament measured one inch and a half in length, and was coiled round the complete worm. On transferring it to a watch-glass containing water, hundreds of embryos made their escape. Owing to the transparency of the tissues, I had much difficulty in finding the reproductive outlet in the perfect worm, and the effort to find the opening was all the greater because my interpretation of Dr. Bancroft's figure had put me off the right track. It seemed so natural to suppose that the 'loops' protruding from the centre of the body resulted from an ordinary prolapsus uteri, so common in preserved specimens of nematodes. At length I found the vagina and its orifice to be situated close to the head (about $\frac{1}{20}''$ from it), the anal orifice being placed within the $\frac{1}{90}$ of an inch from the extremity of the tail. Pl. LXXXVI, fig. 2, *b*. Presumably, these openings were both on the ventral line, but I could not determine the point with certainty. The vaginal pouch, $\frac{1}{100}''$ long, was crowded with embryos, and a constriction marked its junction with the uterus proper, which appeared to me to divide lower down at a distance of $\frac{1}{10}''$ from the head. Towards the tail a fold of the tuba Fallopii was seen to extend to within $\frac{1}{20}''$ of the extremity. All sections of the uterine system were crowded with germs, eggs, and embryos in their usual relative situations.

"My examinations of the ova and embryos were chiefly made from the 'sediment' sent in the special glass tube. The fully formed embryos were $\frac{1}{125}''$ in length by $\frac{1}{2500}''$ in breadth. They each showed a double skin, the outer envelope in the more advanced specimens leaving clear spaces at either end of the body, resulting from commencing ecdysis. Pl. LXXXVI, fig. 9. I saw no trace of intestinal tube, but a central line of condensation marked an early differentiation of the somatic granular contents. The less advanced embryos were mostly enclosed in a chorional envelope, fig. 4, *c*, the smallest free embryos measuring only $\frac{1}{200}''$ in length, by $\frac{1}{3500}''$ in breadth. These had no double contour. The ova, whose yelk-contents were still in various stages of cleavage, gave an average long diameter of $\frac{1}{900}$ to $\frac{1}{1000}$ of an inch, fig. 4, *d*.

"Such are the facts that I have been able to make out. If they do not supply all that one could desire, they, nevertheless, enable me to extend and amend the characters of the species as follows:—Filaria Bancrofti (mihi). Body capillary, smooth, uniform in thickness. Head with a simple circular mouth, destitute of papillæ. Neck narrow, about one-third of the width of the body. Tail of female simple,

Fig. 2.

Fig. 1.

Fig. 3.

Fig. 4.

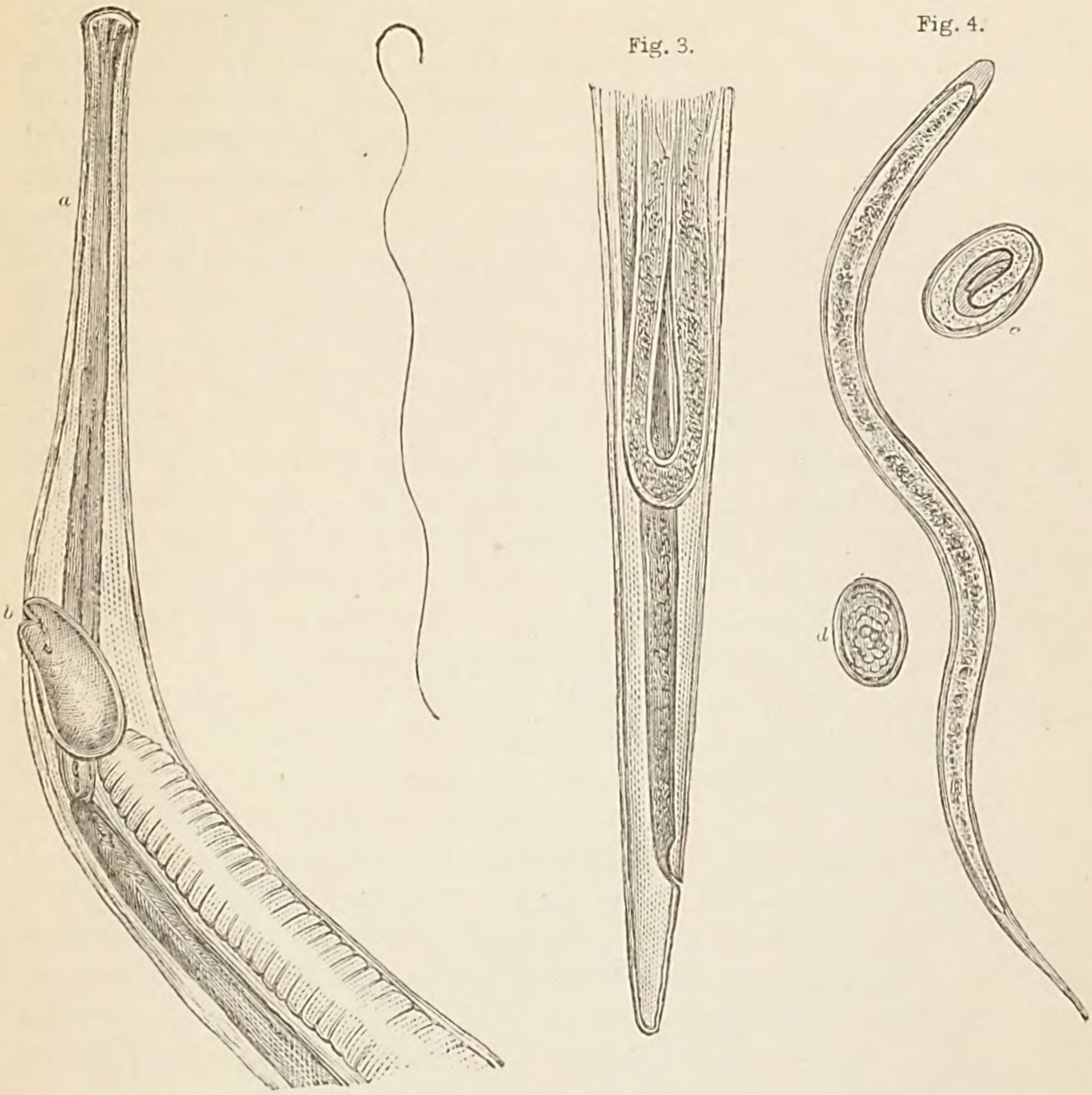
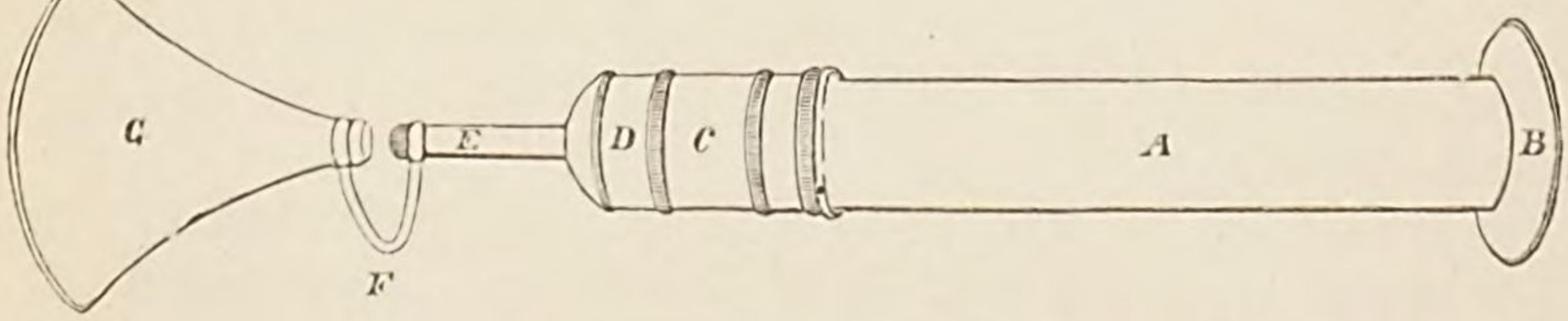


Fig 1, Filaria Bancrofti, female, natural size. Fig. 2, Head and neck, showing œsophagus (*a*) and vagina (*b*). × 55. Fig 3, Tail of the same, showing fold of tuba and termination of intestine. × 55. Fig. 4, Free embryo × 400. (*c*) Intra chorional embryo. × 300. (*d*) Egg. × 360. p. 505. After Dr. Cobbold. *Lancet, October 6th*, 1877.

Fig. 5.



Dr. Urban Pritchard's microscope for examining the circulation in the frænum of the tongue of the human subject. A, Body; B, Eye-piece; and C, Objective; D, a cap fitting over objective, with a fine tube (E) projecting from its centre; F, Connector, allowing space for the frænum of the tongue; G, Speculum for throwing a strong light upon the object. p. 503.

[To face page 504.

bluntly pointed; reproductive outlet close to the head; anus immediately above the tip of the tail. Length of female, $3\frac{1}{2}$ in.; breadth, $\frac{1}{90}''$; embryos, $\frac{1}{200}''$ to $\frac{1}{125}''$ in length, by $\frac{1}{3000}''$ to $\frac{1}{2250}''$ in breadth; eggs, $\frac{1}{1000}''$ by $\frac{1}{1650}''$.

"The male of this worm I have not seen.

"As regards the nomenclature, I have associated Dr. Bancroft's name with the sexually mature worm as being in harmony with the binomial method, and little calculated to mislead. At the same time it helps to fix both the source and date of the discovery (Brisbane, Dec. 21st, 1876).* This concession in the matter of nomenclature, towards a highly meritorious observer, and able surgeon, detracts nothing from the higher merits of Lewis, who first named the immature worm, Filaria sanguinis hominis. Both Dr. Salisbury and myself had previously been made acquainted with intra-chorional embryos, which are, I think, the young of Filaria Bancrofti; but it was reserved for Lewis to discover the hæmatozoal character of the young of this worm and actually to take them from the blood. Should my determination of the generic relationship of these embryos with Filaria Bancrofti be subsequently verified, it would obviously be absurd to call the adult worm Trichina cystica; yet Salisbury gave this name to the urinary parasite. It certainly was a very singular thing that when I was actually treating my little African patient for trematode hæmatozoa, it never once occurred to me that the numerous nematoid embryos, mixed with the Bilharzia ova, were also hæmatozoal. It was alleged that my patient had passed worms two or three inches long by the urethra. Naturally I concluded that these were the parents of the eggs and embryos, and therefore urinary. The inference was wrong; but it has since become instructive, as showing how near one may go towards a discovery without actually making it.

"*Appendix.*—Since the above was written Dr. Lewis has himself furnished additional means of identification. His mature Filaria sanguinis hominis (1874) and my F. Bancrofti (1877) are clearly the same species. I will go further, and express the opinion that all the various larval forms severally described by Salisbury, Lewis, Sonsino, Wücherer, Crévaux, and Corre, Silva-Lima, Bancroft, and myself, are referable to one and the same species."

* Dr. Cobbold seems not to be aware that in his work on "The Pathological significance of Nematode Hæmatozoa," published in 1874, Dr. Lewis gives an account of the mature "Filaria sanguinolenta," with drawings of the male as well as of the female worm.

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THE END.

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